PB83-251298

Potential for Biological Controls of ´Cladophora glomerata´

glomerata´

Academy of Natural Sciences of Philadelphia, PA

Prepared for

Environmental Research Lab.-Duluth, MN

Aug 83

U.S. Department of Commerce
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TECHNICAL REPORT DATA

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1. REPORT NO. EPA-600/3-83-065	2.	3. RECIPIENT'S ACCESSION NO. PB83-251298
4. TITLE AND SUBTITLE The Potential for Biological Controls of *Cladophora glomerata*		5. REPORT DATE August 1983
		6. PERFORMING ORGANIZATION CODE
7. AUTHOR(S) Ruth Patrick, Charles F. Rhyne, R. William Richardson, III, Richard A. Larson, Thomas L. Bott, and Kurt Rogenmuser		8. PERFORMING ORGANIZATION REPORT NO.
9. PERFORMING ORGANIZATION NAME AND ADDRESS Academy of Natural Sciences of Philadelphia Philadelphia, Pennsylvania 19103		10. PROGRAM ELEMENT NO.
		11. CONTRACT/GRANT NO. Grant R-805106
12. SPONSORING AGENCY NAME AND ADDRESS U.S. Environmental Protection Agency Environmental Research Laboratory-Duluth 6201 Congdon Boulevard Duluth, Minnesota 55804		13. TYPE OF REPORT AND PERIOD COVERED
		14. SPONSORING AGENCY CODE EPA/600/03

15. SUPPLEMENTARY NOTES

16. ABSTRACT

The purpose of this research program was to determine whether or not there were natural biological controls of *Cladophora glomerata* that could be developed. Two avenues of research were pursued. One was to study macroscopic organisms that were known to occur in the Great Lakes area to see if any of them would prefer *C. glomerata* as a food source and under what conditions the desirability of *C. glomerata* could be increased. The second approach was to study microorganisms, particularly fungi, which were of common occurrence in aquatic ecosystems to determine if they might function as controls for *C. glomerata*.

The results of these studies show that *C. glomerata* was a poor food for *Physa heterostropha*, *Orconectes propinquus*, *Rana pipiens*, *Ictalurus punctatus*, and *Pimephales promelas*. Not only would these species not eat the food, but if they did eat the food there seemed to be a deleterious effect. In the studies with snails it was evident that egg production was greatly curtailed on the *C. glomerata* diet.

The unialgal *C. glomerata* was less preferred than the *C. glomerata* with epiphytes. The effects on the organisms other than *Physa heterostropha* were mainly losses in weight. The crayfish did not molt even when the eyestalks were removed, and the frog tadpoles did not show any signs of metamorphosing. All organisms steadily lost weight during the experiments.

An examination of the chemical constituents of cells of *C. glomerata* and of diatoms showed that they differed considerably from diatom-dominated periphyton, which was the preferred diet in all experiments. These differences were mainly that the diatoms had larger amounts of amino acids, such as serine, aspartic acid, and glutamic acid. Also the free and combined fatty acids as diatoms were different than those of *Cladophora*. The diatoms were dominated by C_{16} and C_{20} combined fatty acids, whereas the unsaturated C_{18} fatty acid, particularly linolenic acid, was common in *C. glomerata*. Of the free fatty acids the diatom-dominated periphyton contained mainly C_{14} and C_{16} isomers, while *C. glomerata* contained mostly C_{18}. It was interesting to note that of the short-chain fatty acids, *C. glomerata* contained C_{12} acids, particularly lauric acid, and this was absent from the periphyton. It is known from other experiments that lauric acid is toxic to several organisms.

The second avenue of approach was to study the effect of fungi as parasites on *C. glomerata*. One fungus, *Acremonium kiliense* (Fungi Imperfecti) was found to have an antagonistic effect on *C. glomerata*. The effect varied in various experiments; those carried out in the summer months showing the greatest antagonistic effect. It may be that the lesser effect in the fall was due to temperature and other environmental conditions or to the fact that *A. kiliense* had reduced virulence. A toxin was present in the supernatant derived from cultures of *C. glomerata* that had been damaged by this pathogen. It is water soluble and can withstand heat up to 80°C.

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group

18. DISTRIBUTION STATEMENT RELEASE TO PUBLIC	19. SECURITY CLASS *(This Report)* UNCLASSIFIED	21. NO. OF PAGES 194
	20. SECURITY CLASS *(This page)* UNCLASSIFIED	22. PRICE

EPA Form 2220-1 (Rev. 4-77) PREVIOUS EDITION IS OBSOLETE

PB83-251298

EPA-600/3-83-065
August 1983

THE POTENTIAL FOR BIOLOGICAL CONTROLS OF *Cladophora glomerata*

by

Ruth Patrick, Charles F. Rhyne, R. William Richardson, III,
Richard A. Larson, Thomas L. Bott, and Kurt Rogenmuser
Academy of Natural Sciences of Philadelphia
Philadelphia, PA 19103

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EPA Project Officer
Nelson Thomas
Grosse Ile Laboratory
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U.S. ENVIRONMENTAL PROTECTION AGENCY
DULUTH, MN 55804

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ABSTRACT

The purpose of this research program sponsored under Grant R-805106 of EPA was to determine whether or not there were natural biological controls of *Cladophora glomerata* that could be developed. Two avenues of research were pursued. One was to study macroscopic organisms that were known to occur in the Great Lakes area to see if any of them would prefer *C. glomerata* as a food source and under what conditions the desirability of *C. glomerata* could be increased. The second approach was to study microorganisms, particularly fungi, which were of common occurrence in aquatic ecosystems.

The results of these studies show that *C. glomerata* was a poor food for *Physa heterostropha*, *Orconectes propinquus*, *Rana pipiens*, *Ictalurus punctatus*, and *Pimephales promelas*. Not only would these species not eat the food, but if they did eat the food there seemed to be a deleterious effect. In the studies with snails it was evident that egg production was greatly curtailed on the *C. glomerata* diet.

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The second avenue of approach was to study the effect of fungi as parasites on *C. glomerata*. One fungus, *Acremonium kiliense* (Fungi Imperfecti) was found to have an antagonistic effect on *C. glomerata*. The effect varied in various experiments; those carried out in the summer months showing the

greatest antagonistic effect. It may be that the lesser effect in the fall was due to temperature and other environmental conditions or to the fact that A. kiliense had reduced virulence. This toxin is present in the supernatant derived from cultures of C. glomerata that had been damaged by this pathogen. It is water soluble and can withstand heat up to 80°C.

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ACKNOWLEDGEMENTS

This research was made possible by grant #R-805106 from the Environmental Protection Agency. We acknowledge with thanks the constructive help of our project officer, Dr. Nelson Thomas.

The authors wish to express their appreciation to Dr. Raymond Jezerinac, who provided the crayfish for the study; Dr. E. M. Zipf, who aided in the removal of crayfish eyestalks for the molting studies, and Dr. Walter Gams of the Institut fur Schimmelspilze, Baarn, the Netherlands, who confirmed our generic identification of *Acremonium kiliense*. Mrs. Lee Anderson has carried out the editing and preparation of the manuscript.

SECTION

EXECUTIVE SUMMARY

In this research two avenues were followed in trying to identify a biological control of *Cladophora glomerata*. One of these was to study macroorganisms common to the Great Lakes region which previous research has shown select algae as a considerable part, if not the main part, of their diets. The other avenue of research was to find a microorganism which by parasitism would destroy *C. glomerata*. It was realized that this organism must be fairly specific for *C. glomerata*, and if present in fairly large amounts would not destroy other algae that were desirable food sources.

Studies with Macroorganisms

The organisms selected for the macroorganism studies were the pond snail, *Physa heterostropha*; the crayfish, *Orconectes propinquus*; the tadpole, *Rana pipiens*; the channel catfish, *Ictalurus punctatus*; and the fathead minnow, *Pimephales promelas*.

All of these organisms feed more or less extensively on algae. We did not want to use any exotic species to the Great Lakes, as these might pose other problems if introduced. Some of these, such as *Tilapia aurea*, are known to be prodigious algal feeders. In these experiments most of the work was done on *Physa heterostropha*, *Orconectes propinquus*, and *Pimephales promelas*.

All of the studies showed that *Cladophora glomerata* was not a satisfactory diet. Diatom-dominated periphyton was a more satisfactory diet, as the organisms had a better rate of growth on that diet than on *Cladophora* plus epiphytes. The *Cladophora* plus epiphytes tended to be a better food source than unialgal *Cladophora*. In the experiments using *Physa heterostropha* some snails were starved. Their decline in weight and their respiration rates tended to be greater when starved than when fed unialgal *C. glomerata*.

The results of the experiments with *Physa heterostropha* showed that the relative value of the three diets presented were diatom-dominated periphyton > *Cladophora* plus epiphytes > unialgal *Cladophora*. The respiration rates of snails fed *Cladophora* or *Cladophora* plus epiphytes were less than those on the periphyton diet. There was not much difference between diets of *Cladophora* plus epiphytes and unialgal *Cladophora*.

The reproductive success was much greater on the diatom-dominated periphyton diet and was very poor on the unialgal Cladophora diet. Egg-laying greatly decreased in a relatively few days (11-15 days) after the experiments started. However, the eggs produced from all three diets had similar caloric content. These experiments were repeated at various seasons of the year and showed similar results.

The experiment with Orconectes propinquus showed that none of the diets were very satisfactory, but that the diatom-dominated periphyton was much better than the other two diets. The crayfish molted for only a short time after the experiment started on July 28. This may be due to the normal life cycle trends as crayfish tend to hibernate in the cooler months. However, the temperature at which the experiments were run was similar to summer temperatures. Molting was forced by removing the eyestalks and associated X organ-sinus gland. Following this operation in January the females on the periphyton diet all molted.

The respiration rates on all three diets declined during the experiments, and the rates of the females tended to be greater than the males.

The experiments with the tadpoles of Rana pipiens resulted in increases in size, and there were signs of metamorphosis on the diatom-dominated periphyton diet and on Cladophora plus epiphytes diet. Those fed on unialgal Cladophora grew very little and showed no signs of metamorphosis. The respiration rates for tadpoles were similar on the periphyton and Cladophora plus epiphytes diets, and were much lower for tadpoles on the Cladophora diet. These tadpoles were very small and weighed very little, as they had lost weight.

The experiments with the channel catfish (Ictalurus punctatus) showed a similar pattern. The diatom-dominated diet was more satisfactory than the unialgal Cladophora diet. However, when compared with the fish fed on prepared fish food, none of the diets were as satisfactory.

The fathead minnow (Pimephales promelas) ingested and assimilated much more of the diatom-dominated periphyton than the Cladophora plus epiphytes or the unialgal Cladophora. The fish fed the last two diets decreased in weight, with those on unialgal Cladophora showing the greatest decrease. The respiration rates were variable for fish on the periphyton diet. It was highest for those fed on Cladophora plus epiphytes. On both the Cladophora diets the cal/gm/hr (Table 32) were higher than on the periphyton diet.

These results led us to an examination of the relative food value of Cladophora glomerata and diatom-dominated periphyton. Determination of the calories/mg showed similar amounts.

An analysis of the chemical composition of diatoms versus Cladophora showed some interesting results. The amino acids of diatoms that were in larger amounts were serine, aspartic acid, and glutamic acid, and a small amount of cystine which was absent in *C. glomerata*.

The combined fatty acids in diatoms were dominated by C_{16} and C_{20} acids. Unsaturated C_{18} fatty acids, particularly linolenic acid, was much lower in diatoms. *C. glomerata* was more like green plants, having C_{18} unsaturated fatty acids, particularly linolenic acid, and low concentrations of C_{16} and C_{12} acids.

Periphyton contained more free fatty acids than *C. glomerata*, with the majority being C_{14} and C_{16} isomers, while those present in *Cladophora* were mostly C_{18}.

Of the short chain free fatty acids *Cladophora* contained C_{12} (lauric acid), which was absent in periphyton, and C_{10} (capric acid) was in periphyton but not in *Cladophora*. Lauric acid has been shown from many reports to be toxic.

Experiments with yeasts (*Saccharomyces cervisiae*) showed that the extract of *Cladophora* containing this acid was toxic to yeast. This suggests that the presence of this acid may be in part the reason why *C. glomerata* was an unsatisfactory food.

As to soluble carbohydrates, *C. glomerata* contained 4.5%, whereas the periphyton contained 21.6%. There was also more phosphorus in the periphyton.

The results of the studies show that there are several differences in the chemical composition of the periphyton dominated by diatoms and unialgal *C. glomerata*. These differences would result in differences in nutrient value of these two food sources. In *C. glomerata* lauric acid was present, which may be toxic to the organisms studied. Further research is needed to verify this potential toxicity.

Studies with Microorganisms

The second avenue of approach was to determine whether fungi commonly occurring in the aquatic environment would have an antagonistic effect on *C. glomerata*. One isolate, *Acremonium kiliense* (Fungi Imperfecti) was found to have an antagonistic effect on *C. glomerata*. This fungus was isolated from the stock culture of *Cladophora* where it was present in small amounts. This fungus in flask cultures deleteriously affected the cell condition of *C. glomerata* and caused a reduction in chlorophyll *a* when the stock cultures of *Cladophora* were used. *Cladophora* that was isolated from Lititz Creek was more susceptible to the fungus than *Cladophora* from stock cultures. In the Lititz Creek community experiments there was not as good a correlation of loss of chlorophyll *a* with damage to the cells. This may be due to the fact that small amounts of algae were present that were not damaged by *A. kiliense* and thus would obscure the loss of chlorophyll *a*. Therefore, it was decided that direct microscopic examination of the cells was the most reliable way to determine the damage of this fungus.

Supernatant culture medium from the flasks where *Cladophora* had been damaged when added to other undamaged *Cladophora* cells also had deleterious effects on the algae. This antagonistic substance from *A. kiliense* is water soluble and does not seem to be damaged by heating to 80°C. Cultures of *A. kiliense* were raised on Czapek-Dox broth, and *Cladophora* was inoculated with conidia, but these did not produce damage.

Microcosm experiments were then set up in which the *C. glomerata*-dominated communites were exposed to varying concentrations of *A. kiliense*. The first experiment was run from July 17 to August 1, 1978, and the second was run from August 11 to August 31, 1978. In both cases there was a striking effect of these fungi on the *Cladophora*, although diatoms and *Ulothrix* living in the microcosms were unaffected. In fact, the *Ulothrix* divided rapidly and occupied the habitats formerly occupied by *C. glomerata*. *Scenedesmus* and *Spirogyra* were also present, as were protozoans and rotifers, in the experiment carried out August 11-31. They did not seem to be affected by this fungus. Static microcosms produced similar results.

These experiments were then repeated in the fall of the year during the period from September 18-October 4. In these experiments the effect was not as apparent. The experiment was again repeated October 10-20 in a flowing stream microcosm, and although there was some effect it was not nearly as dramatic as in August. It may have been the difference in day length, in temperature, or a variety of environmental conditions that reduced the antagonistic effect of the fungus. It also may be that the fungus in culture loses its virulence, and therefore is not as effective as freshly isolated cultures.

More studies need to be carried out to determine just what the chemical is that is causing this effect, whether the fungus loses its virulence on culturing, and/or what other environmental conditions may affect its virulence. An experiment was run toward the end of the study using 100-400 mg/l of cephalosporin. This produced some chlorosis, but in concentrations of 10-50 mg/l had no effect. Whether this is the compound that produced the antagonistic effect in the previous experiments is not known, and further studies need to be carried out.

From these various types of studies it would appear that microscopic organisms have greater promise as effective biological controls than the larger organisms.

SECTION 1

INTRODUCTION

The purpose of this research program was to determine if biological controls of the growth of Cladophora glomerata could be developed. This alga is becoming a nuisance growth in many of the Great Lakes. It is also a nuisance growth in many of the rivers of the United States. This alga belongs to the Family Cladophoraceae, Order Cladophorales, in the Division Chlorophyta.

Two methods of approach have been used in trying to answer this question. One was an attempt to identify one of the naturally occurring species in the Great Lakes that would preferentially feed on Cladophora glomerata. In other words, are there species in this area for which C. glomerata is a satisfactory food?

The second approach was to determine if there was a microbial parasite naturally occurring in the area or commonly occurring in natural environments that if encouraged could become a control of this alga.

The need to determine the chemical characteristics of C. glomerata became evident during the course of these experiments.

Since the procedures and methods of study were so different for these two approaches to the problem, the report is divided into three sections for discussion. The executive summary contains a general discussion of these sections.

The three sections are:

I. The potential of Cladophora glomerata as a satisfactory food for various invertebrates and fish, by Ruth Patrick, Charles F. Rhyne, and R. William Richardson, III.

II. Chemical composition of Cladophora glomerata cells, by Richard A. Larson

III. Fungal pathogen for possible biological control of Cladophora glomerata, by Thomas L. Bott and Kurt Rogenmuser

SECTION 3

RECOMMENDATIONS

For developing a biological control the results of this research indicate that the fungus *Acremonium kiliense* may be a potential biological control. However, a good deal more experimentation has to be done to understand the conditions under which it develops and what the toxin is to *Cladophora glomerata*. The fact that it does not seem to damage diatoms, *Ulothrix*, *Scenedesmus*, *Spirogyra*, protozoa, nor rotifers indicates that it may be confined in its effects to a relatively few species.

The results from the experiments with megaorganisms as biological controls of *C. glomerata* produced negative results. However, they did turn up some very interesting information as to the nutritional value of *C. glomerata*. This alga has a very different assortment of free and combined free and fatty acids and different amino acid proportions than diatom-dominated periphyton, which was obviously the preferred diet. It also contains C_{12} (lauric acid) which has been shown to be toxic to some aquatic organisms.

Further studies should be carried out to determine whether it is the lauric acid that is producing the effect that inhibits the eating of this algae by various megainvertebrates and whether it also produces a toxic effect on the organisms. An attempt should be made to determine its mode of action--that is, whether membranes or damaged, enzyme systems inhibited, etc. If further research clearly shows that this alga inhibits the reproduction of many snail species, particularly those that are vectors of schistosomiasis, this alga might be used as a control mechanism for this disease. *C. glomerata* is a very tolerant alga and can grow under a great variety of conditions. The question also of interest is whether other algae that develop large populations under eutrophic conditions contain toxic substances.

I. THE POTENTIAL OF CLADOPHORA GLOMERATA AS A SATISFACTORY FOOD FOR VARIOUS INVERTEBRATES AND FISH

INTRODUCTION

In order to determine whether *Cladophora glomerata* is a satisfactory food for a predator, the growth and, if possible in the time allotted, the reproductive success of the species fed only *Cladophora* should be compared with other available algal diets. It was noted that *C. glomerata* in nature has many epiphytes growing on the filaments, and most of these are diatoms. Therefore it was decided that the three food sources to be studied were a mixed periphyton dominated by diatoms, *C. glomerata* with epiphytes, and unialgal *C. glomerata*. We had hoped to raise axenic cultures of the alga but as pointed out by Gerloff and Fitzgerald (1) and others, no one has succeeded in doing this.

The naturally occurring species selected for study were as follows. The snail, *Physa heterostropha*, was chosen because it is a widely distributed pulmonate and can live in water of varying oxygen concentrations. It has been found (2) to have a toxicity tolerance intermediate between diatoms and bluegill sunfish, *Leopmis macrochirus*, and it is an excellent food for many fish. The crayfish, *Orconectes propinquus*, was chosen because it is native to the Great Lakes area and is an important part of the food web in the area. Tadpoles, *Rana pipiens*, were chosen for a similar reason. The fathead minnow, *Pimephales promelas*, was chosen because it is commonly occurring in this area, and because its general physiology is well known through the work of Mount, Brungs, and others. The tests on channel catfish, *Ictalurus punctatus*, were to determine if this fish would eat and grow on this food, and were only carried out for a short period of time. The most extensive tests have been on *P. heterostropha*, *O. propinquus*, and *P. promelas*. Time did not allow more extensive tests on the other organisms.

REVIEW OF PERTINENT LITERATURE

Snails

There are numerous articles in the literature concerning the effects of various types of algal and invertebrate foods on the growth and reproduction of various pulmonate molluscs. Bovbjerg (3) studied the food preferences of the snail Stagnicola reflexa Say. He found that the snail preferred the alga Spirogyra sp. over the higher plants Ceratophyllum demersum, Myriophyllum exalbescens, and Ranunculus longirostris. He found that from a distance of 1-2 cm the snails did not perceive this food by any chemoreception. Only when the snail physically encountered the food it preferred would it stop and eat, otherwise it would continue to move. In other studies with different plants the snail moved three times as much on green and dead cattails, bladderwort, buttercups, liverworts, and duckweed than it did on the algal food.

Bovbjerg (3) in his studies with the lymnaeid snails Stagnicola reflexa, S. exilis, S. elodea, and Lymnaea stagnalis, found that these snails preferred pellets made from crayfish over those made from plant food and that they could detect from some distance the pellets of crayfish, whereas they could not detect those of plant food. He found there was also little response to the plant filtrate of the pondweed, Potamogeton richardsonii. For growth the best diet was a mixture of plant and animal material, and only on this mixed diet did the animals reproduce.

Calow (4) studied the snail, Lymnaea pereger, to determine its preference for various types of food sources. This snail is commonly found growing on Elodea. An examination of the guts showed that some of the Elodea tissue was in the gut, but that it was mostly filled with epiphytes. He incorporated this in an agar block, and in other agar blocks he incorporated singly crushed Elodea, mixed diatoms, and a mixture of filamentous algae. From this study he found that the greatest preference was for the epiphytes taken from the Elodea and for the filamentous algae. He states that the work of Paine and Vedes (5) shows that the ash-free dry weight of the diatoms have the greatest potential energy stored--that is, the kcal/g ash-free dry weight is greater in diatoms than in the other algae. The reason why this snail seemed to prefer the mixed culture of algae was that it was easier to eat, there being a harder cell wall about the Elodea and silicious walls on the diatoms. He states that the preference of this snail was first for the filamentous algae and the extract of epiphytes from the Elodea, secondly for the diatoms, and thirdly for the Elodea.

Eisenberg (6), working with the pond snail Lymnaea elodes, found that the amount of food is of major importance in determining the growth and fecundity of this snail. Growth, weight, and total eggs were strongly correlated with the food supply. The eggs/egg mass did not change very much with food supply, but did increase somewhat. The total number of egg masses, however, increased greatly with food supply.

Hunter (7), working with Lymnaea palustris, determined that the stage of the life history was very important in regard to the physiological condition of the snail. He found that this snail had the greatest amount of nitrogen and carbon (ug/snail) at the mid-breeding period, and that there was considerable variation from snail to snail in the amount of these elements. The mean dry weight was greatest at this period. He placed this snail in two different environments--one designated as MAR and the other as FAY. He found that the snails that typically grew in the MAR environment when placed in the FAY environment, and those snails that typically grew in the FAY environment were much more fecund in the FAY environment. He did not state just what the characteristics of the environment were that he believed produced this.effect.

Studies of Laevapex fuscus and Lymnaea palustris indicate that they prefer an intermediate carbon content diet which is high in nitrogen and probably protein, and this is best for their growth and reproduction. Diatoms, bacteria, and blue-green algae have a high nitrogen content. McMahon, et al. (8) state that the qualities of food are more important than the quantities.

Elwood and Goldstein (9), working with Goniobasis clavaeformis, examined the ^{32}P which was sorbed onto the periphyton and then ingested by the snail. He found that the food ingestion increased with increased temperature up to 14°C. The elimination of absorbed phosphorus also increased with increased temperature, so that the shortest retention time was 6 days at 19.3°C as compared with 34 days at 10°C. The absorption efficiency was fairly constant and was about 39%; they speculate that perhaps the efficiency may be related to gut clearance.

Paine and Vedes (5) studied the relative food value of green, red, and brown algae and the diatom Nitzschia paradoxa. They found that the broad-leaved algae, such as Laminaria macrocystis and Gigartina sp. were the most preferred foods and had intermediate food value. They found the highest food value in Nitzschia paradoxa, which was 5.47 kcal/gm ash-free dry weight and the caloric value in the marine form of Cladophora sp. was highly variable and averaged about 5.17 Kcal/gm ash-free dry weight.

Spight and Emlin (10) studied two marine snails, Thais lamellosa which spawns annually and T. emarginata which spawns many times during the year. They found that growth was correlated with food supply, and that clutch size increased exponentially with shell length. For example, *T. emarginata* triples its fecundity as it moves from minimum mature size to average mature size; whereas *T. lamellosa* doubles its fecundity as it moves from minimum mature size to average mature size. In the larger animals there are larger capsules or egg masses produced and more eggs in both species. It is interesting to note that *T. lamellosa*, which reproduces once a year, produces about the same number of eggs as *T. emarginata* which spawns many times during the year.

Nicotri (11) studied the diets of the marine molluscs *Collisella pelta*, *C. strigatella*, *Notoacmaea scutum*, and *Littorina scutulata*. He found that all of these snails selectively removed and seemed to prefer three species of diatoms. These were *Melosira moniliformis*, *M. nummuloides*, and *Fragilaria striatula* var. *californica*. These were among the largest and most common diatoms. He found a great many frustules of *Nitzschia frustulum* in the guts of these organisms, but he felt that because of their small size they probably did not contribute much to the diet. He found that *L. scutulata* was less efficient at digesting *Achnanthes brevipes* and *Synedra tabulata*.

Runham (12) points out that as yet the nutritional requirements of pulmonate snails are virtually unknown. We do not know, for example, their requirements for proteins, fats, carbohydrates, vitamins, and minerals. Some species such as *Limax flavus* (L.) can live completely on wheat flour; however, the growth rate and size were reduced. For some snails the requirements of vitamins have been demonstrated. *Biomphalaria glabratum*, when reared under axenic conditions and fed on a diet of *Escherichia coli* and yeast, grew well but did not lay eggs unless vitamin E was added to the food (Runham, quoting Vieira, 12). Runham also pointed out that to be a suitable food the texture of the cell wall, its smell, and its taste are important as well as its caloric value. Thus we find a great variation in the kinds of preferred foods of this group of organisms.

Crayfish

Crayfish are known to be scavengers and to feed on a wide variety of foods. Typically they are detritus feeders and seem to be more or less carnivorous. However, Budd, et al. (13) have shown that they could maintain a breeding population of crayfish by supplying them with a rich green-algal flora. Bovbjerg (14) determined that when the crayfish become independent of the yolk sack at the time of the fourth instar, they are filter feeders and feed upon algae.

Magnuson, et al. (15) summarized the data on the ability of various species of crayfish to control aquatic vegetation. All of this vegetation is higher plants, and no control of algae by the use of crayfish seems to be evident in the literature, except for *Chara* sp. and *Nitella* sp. It has been shown that Orconectes rusticus will control Potamogeton, Vallisenaria, Ceratophyllum, Myriophyllum, Megalodonta, Nuphar, and Nitella. It should be pointed out that the introduction of crayfish for the control of macrophytes may produce a bad side effect in that they are known to feed upon the eggs of several species of fish.

In studies conducted by Momot, et al. (16), it was evident that the crayfish Orconectes propinquus and O. virilis fed largely on the benthos, and the algae was a large part of this benthos. Analyses of the stomachs of the young-of-the-year of O. virilis indicate that the highest amount of any one food was plant detritus that occurred in all the stomachs examined. The next highest was pine pollen that occurred in 82.9% of the stomachs examined, and diatoms that occurred in 45.1% of the stomachs analyzed. Twenty-nine percent was in the form of chironomid eggs, and 25.6% was fragments of arthropods. These data indicate that the crayfish O. virilis is an omnivore feeding on many different kinds of organisms and that the type of organism it seems to prefer is the benthos.

Momot (17) found that O. virilis was primarily an herbivore in marl lakes, feeding chiefly on algae and the "aufwuchs" with marl incrustations. In the marl lake, West Lost Lake, the biomass of crayfish was nearly 10-fold that of other invertebrates, and it was believed that this was due to their ability to utilize algae.

The crayfish O. virilis usually goes into hibernation about the first of October. The maturation of eggs takes place during the winter months. This maturation process is affected by the X organ-sinus gland complex of the eyestalk. Temperature and photoperiod are necessary for the proper maturation of the eggs (18).

The work of Aiken (19) shows that the molt cycle of O. virilis is regulated by environmental factors, especially photoperiod, and by certain endocrines. The endocrine system modifies the inducive effect of the photoperiod. The photoperiod seems to control the interaction of two hormones designated as MH and MIH. Proecdysis (molting) is induced when the influence of MH exceeds that of MIH. If after the onset of proecdysis the titer of MIH interferes with premolt preparation for one or more of these molting steps, the molt attempt is terminated at that point where the affected step occurs. A lesser amount of MIH may simply prolong proecdysis, whereas a larger amount may suspend it indefinitely. The photoperiod necessary for molting to proceed is 15-16 hours for subthreshold lengths in November and December, marginal lengths in January, and clearly above the threshold lengths in February. The MH hormone seems to be produced from the paired endocrine organs in the thorax, whereas the molting-inhibiting hormone MIH is from the X organ-sinus gland complex of the eyestalk.

Fish

Fathead Minnows (Pimephales promelas) - There is a great diversity of opinion as to what is the food of this fish. Smith, et al. (20) noted in his studies that they could grow and reproduce when fed brine shrimp. Held and Peterka (21) state that the main food is Crustacea, particularly Cladocera, and that algae furnish only a small percent of the ingested food (0.2% by volume were mainly diatoms). Lord (22) stated that these fish are mainly algal feeders and that they feed upon the filamentous green algae, Oedogonium and Spirogyra, and probably other filamentous algae, which he does not name. They also eat a great many diatoms. Coyle (23) noted that fish ranging in size from 25-40 mm feed upon diatom slime and small algae, which they nibble off the stems of submerged and emergent aquatic plants.

Channel Catfish (Ictalurus punctatus) - The food habits of blue and channel catfish have been studied. The channel catfish feeds primarily upon amphipods, small insects, algae, and various types of organic matter (24). Devaraj (25), in his studies of Ictalurus punctatus, found that 47.7% of their food were dipteran larvae, and that the algae were 1-23% of the gut content. These were relatively small fish. When they became larger--i.e., 300 mm long--fish formed about 87% of the volume of food in the gut, according to Ambrose and Brown (26, cited by Devaraj, 25). Perry (24) found that the channel catfish up to a length of 376 mm fed primarily on amphipods, small insects, algae, and undetermined organic material.

Similar results as to the value of the blue-green algae as compared with the green alga Spirogyra sp. were found by Stanley and Jones (27) in their studies with the bigmouth buffalo, Ictiobus cyprinellus, and Tilapia aurea. They found that the conversion rate when these fish were fed on blue-green algae was 2, whereas when the grass carp, Ctenopharyngodon idella, was fed on Spirogyra sp. the conversion rate was about 10 and poor growth resulted.

Repsys, et al. (28), in their studies of the black bullhead, Ictalurus melas, found that 32% of the food in the gut was planktonic Cruatacea, 25% chironomids, 27% fish, 9% crayfish, 2% filamentous algae, and 5% miscellaneous organisms. However, in the young bullhead 94.4% was planktonic Crustacea and 5.6% was chironomids. No algae were noted.

The results of feeding studies on the brown bullhead, Ictalurus nebulosus, indicate that 23% of the carbon presented as Spirogyra sp. and 67% of the carbon presented as Anabaena flos-aquae was assimilated by this fish in

24 hours. These data indicate that *Anabaena flos-aquae* is a better food source than *Spirogyra* (29).

Tadpoles

Various tadpoles have been studied as to their preferred food. It is well known that tadpoles eat largely algae before they metamorphose and develop their hind legs. Jenssen (30) found that the green frog tadpole, *Rana clamitans*, ingested algae in proportion to their number in the environment. He found that they ingested *Pithophora* and *Cladophora*. However, Kamat (31) found that they ingested algae in proportion to its frequency in the pond water but that they did not ingest *Pithophora*, *Cladophora*, and *Chara*. Thus there seems to be a difference in the results of these two studies.

Richards (32) found that algal-like cells inhibited the growth of *Rana pipiens*. The algal cells were not positively identified and may belong to the genus *Prototheca*. These cells seemed to enlarge in the gut of the tadpole and when passed in the feces divide in the water. In concentrations of $1 \times 10^6 - 4 \times 10^7$ cells in the tadpoles' environment, they will inhibit growth.

Sin and Gavrila (33) in the studies of the tadpoles of *Rana ridibunda*, found that the main food was aquatic macrophytes and secondarily food of animal origin.

Dickman (34) found that the tadpoles in Marion Lake, British Columbia, fed largely on *Mougeotia*, a filamentous green alga, shortly after they hatched from the eggs. Thus their predation greatly altered the algae forming the periphyton.

Franz (35) studied the feeding habits of the larvae of the tailed frog, *Ascaphus truei*, from two streams in northwestern Montana. Coyle Creek was dominated by diatoms. The analyses of the guts of tadpoles from this stream showed them to consist primarily of diatoms. In Ward Creek, where *Spirogyra* and *Monostroma* were the dominant algae, examination of the guts of the tadpoles showed that they were filled mostly with diatoms with a few strands of *Ulothrix*. Although *Spirogyra* and *Monostroma* were the most common algae in the stream, no filaments of these algae were found in the guts. He then conducted an experiment placing five larvae into plastic cartons containing a pure culture of *Spirogyra*, another five placed into a carton containing a pure culture of *Monostroma*, and a third group with only *Ulothrix*. After four hours the intestines of the larvae were examined and no algae were found in the tracts of these organisms. Only a few diatoms were present.

METHODS AND PROCEDURES

Obtaining and Culturing of Unialgal Cladophora

Cladophora glomerata was initially obtained from nearly streams. The identification of this species was carefully checked in order to make sure that it was the same species as commonly found in the Great Lakes area.

The isolation of this *C. glomerata* was achieved by allowing filaments to grow up to approximately 1 cm on 1% agar made with the culture medium of Fitzgerald and Gerloff (Medium II, 1). Several filaments were laid out on the surface of the agar and the petri dish was inverted below an artificial light source. In two to three weeks filaments had grown sufficiently so that filament tips could be aseptically removed. These filament tips were placed on fresh agar, and the procedure was repeated several times. The entire procedure was carried out in a hood using aseptic techniques. Finally the unialgal filament tips, free from epiphytes, were transferred to liquid culture Medium II (1). This medium was used throughout all experiments for the growth of *C. glomerata* under unialgal conditions.

The culture vessels used were 4-liter aspirator bottles and 2.8-liter Ferback flasks containing 2 liters and 1.5 liters of culture medium respectively. The cultures were illuminated from below by 40-watt Vita-lite fluorescent tubes, and moderate aeration was continuous. A 15-hour photoperiod at 100 u einsteins/m^2/sec was used, and temperatures were maintained between 21° and 23°C.

Culturing of Periphyton

The periphyton cultures were maintained in a greenhouse by seeding large plexiglas plates (13 cm x 23 cm x 2 mm), with natural periphyton from White Clay Creek, Chester County, Pennsylvania. These were maintained by allowing stream water of suitable velocity to flow continuously over the plates. Within about two weeks the plates were covered with a well-developed periphyton growth. The growths were examined periodically under the microscope and were found to be diatom-dominated, although small amounts of algae and some protozoans occurred from time to time. The dominant diatoms varied seasonally, but *Melosira varians* was generally dominant with smaller populations of *Synedra* spp., *Nitzschia* spp., *Navicula* spp., *Achnanthes* spp., *Cocconeis* spp., and *Gomphonema* spp.

Plates seeded with periphyton were allowed to sit in inclined trays for 20 minutes before weighing in order to remove any excess water. The bottoms of the plates were then wiped clean and dried, and wet weight of the plate plus the periphyton was determined. When the plates were removed after they had been preyed upon, they were dried in a similar manner, and then reweighed. The periphyton was then scraped from the plates and the plates weighed. In this way an estimate of the periphyton consumed was obtained.

Organisms used in the Experiments

Physa hetersostropha - This pulmonate snail was originally collected from nearby sections of Buck Run Creek in East Marlborough Township, Chester County, Pennsylvania. The snails were subsequently reared in the greenhouse. Succeeding generations of these snails, reared on periphyton in a continuous flow-through system, were used in these tests. They were maintained at temperatures of 20° ± 1°C in a 12-hour photoperiod.

Orconectes propinquus - This species of crayfish was collected in Erie and Geauga Counties, Ohio. They were collected by Dr. Raymond Jezerinac. Over 200 individuals were separated by size (eliminating very small and very large crayfish) and by sex. The males were all in form 1 and were on an average larger than the females. All crayfish were put into 550gallon glass aquaria containing rocks which had been previously scrubbed. These were to provide a satisfactory habitat with hiding areas until the experiment started. Males and females were kept in separate tanks.

Pimephales promelas - The fathead minnow was obtained from the Newtown Fish Toxicity Station of EPA in Cincinnati, Ohio. These were roughly cm long when the experiment started.

Rana pipiens - The tadpoles were purchased from the Carolina Biological Supply Company at Burlington, North Carolina. These tadpoles were placed in a large 55-gallon aquarium until the experiments started.

Ictalurus punctatus - The fingerlings of the channel catfish were supplied by the Fish and Wildlife Service at Orangeburg, South Carolina. These fish averaged approximately 3.5 cm in length, and weighed approximately 0.5 gm each when received.

Conditions of Experiments

1. Snail Experiments

The snail experiments were carried out in 20-liter capacity polyethylene trays (18 cm x 45 cm x 23 cm). Two closed recirculating water systems were used in parallel. These were positioned over 90-liter reservoirs. One submersible pump at the bottom of each reservoir delivered water to the trays at the rate of 1-2 liters/min/tray. Water flowing out of each tray was returned by gravity to a reservoir through a loose cellulose filter. Weekly 50-60% of the water in each recirculating system was replaced by fresh stream water at experimental temperatures. The snails being fed on unialgal Cladophora were in one recirculating system, while the snails fed on diatom-dominated periphyton were in the other.

During the winter experiments in which we tested four diets, the setup was somewhat changed. The starvation and unialgal Cladophora diets were in one recirculating system, while the snails being fed Cladophora with epiphytes and those being fed the diatom-dominated periphyton were in the other. Water in the unialgal Cladophora system was initially filtered through a 10-15 u filter, and such filtering was carried out during the weekly water replacements. It was found that such filtering greatly reduced any chance of contamination of the system with other algae.

In the 1977 spring-summer experiments, 27 snails of fairly uniform size that had been reared in the laboratory were placed in each animal tray. Three trays of snails were placed on each of the two diets--unialgal Cladophora and diatom-dominated periphyton. In the 1978 winter experiments 20 snails of fairly uniform size that were reared in the laboratory were placed in each animal tray. Two trays of snails were placed on each of the following diets: unialgal Cladophora , diatom-dominated periphyton, Cladophora with epiphytes, and no food at all. Two arbitrary size classes of snails were used--7-8 mm and 8-9 mm total shell length. Each diet had 20 individuals of each size class in separate trays.

Introduction of Food

In the 1977 experiments food was continually present and renewed as needed to insure that a source of food was never limiting. Cladophora was replaced periodically although very little had been eaten.

Through observation it was learned that these snails typically feed at night. It was therefore decided in the 1978 experiments that food would be introduced at 5:00 one afternoon and removed at 12 noon on the following day. The wet-weighed amounts of periphyton and Cladophora with epiphytes were renewed daily, while the unialgal Cladophora was renewed only twice a week because it was not eaten. At all times more food was present during the feeding periods than could be consumed, and the remaining food was removed and the wet weight determined. These estimates were only made during the 1978 experiments.

The purpose of the feeding procedures in 1977 and 1978 were to make sure that food was not a limiting factor.

The periphyton was introduced as growth on Plexiglas plates, and the snails were allowed to feed directly on the plates. The unialgal Cladophora and the Cladophora with epiphytes were anchored to the bottom of the tray by short glass rods.

Growth Studies

The estimates of growth were made by determining the change in weight of the snails throughout the experiment. To accomplish this a specific number of snails was removed at random initially and periodically throughout the experiment from each test. In those cases where the wet weight was obtained, the snails were dried on a paper towel to remove all loose moisture and then weighed. Following weighing they were returned to the container.

Dry weight was obtained by immersing the snail in boiling water for 15 seconds and extracting the tissue with forceps. The dry weight was obtained by drying at 60°C for 24 hours, and the ash-free dry weight by drying at 500°C for two hours. The caloric values were determined by the use of a Parr Bomb Calorimeter (36).

Respiration Tests

The oxygen uptake was measured at 20°C using a Gilson GRP 20 Differential Respirometer (37). This temperature was used because the average temperature in the test tanks during the summer was 22°-23°C and during the winter was 19.9°C.

Individual snails were taken from each experimental tray and placed in a test vessel with 7 ml of filtered (0.45 u) stream water and acclimated to test conditions for 1 hour. Oxygen consumption was measured every thirty minutes for a 2-3 hour period, and the carbon dioxide that evolved was absorbed by KOH. Similar sized individuals were placed in each vessel, although the number per vessel varied from 1 to 5 depending upon the snail size. This assured that measurable quantities of oxygen would be consumed in each vessel while depletion would not occur prior to the end of the experimental test period. The test animals were free to move in the vessel and had not been previously starved.

Fecundity Studies

Each test tank was examined for egg masses and eggs almost daily except for weekends in the 1976 and 1977 experiments. In the 1978 experiments the egg mass collections were done at less frequent intervals.

Data concerning the number of egg masses and the number of eggs in each mass were recorded only on days after which eggs had been removed from the tanks. By this means one can be sure that the estimates are fairly correct.

Upon removing the egg masses the eggs were examined under the microscope to see if they were viable. A very few instances of non-viable eggs were recorded. Data concerning the caloric content of the eggs were obtained by the use of microbomb calorimetry (38).

2. Crayfish Experiments

The crayfish experiments were carried out in 55-gallon glass aquaria. On the bottom of each aquaria were placed rocks of approximately 3 x 4-5 inches in order to afford a natual habitat and protection from the other crayfish at the time of molting. Twenty crayfish were placed in each aquarium. Each individual was marked in the aerola section of the carapace, using a combination of three different fingernail polishes. No detrimental effects were observed. Three diets were used in these experiments; the same diet was put into two of the aquaria. The Cladophora and periphyton were cultured as described in the snail experiments. The Cladophora with epiphytes was collected from a holding pond at the laboratory and allowed to stand in trays of the stream water for 24 hours. Diurnal temperature increases and low oxygen levels forced the many insect larvae and Planaria out, leaving the Cladophora less contaminated. Upon examination this Cladophora not only contained epiphytic diatoms, as in the case of the snail experiment, but also had associated with it small amounts of Spirogyra and Stigeoclonium.

Introduction of Food

A review of the literature and observations in nature indicated that these crayfish feed mostly at night. Therefore, the food was wet-weighed and introduced at 5:00 p.m. daily and taken out at 8:00 a.m. the following day. During the feeding period the aquaria were without light. The Cladophora was placed on filter paper to drain off all the excess water and the wet weight obtained. In the morning when the Cladophora was removed it was again dried in a similar manner and weighed.

It was found that a considerable amount of the Cladophora was dislodged and broken by the movement of the crayfish. An experiment was conducted to study the amount of Cladophora filament breakage caused by crayfish feeding during 4- and 16-hour periods over a 5-day interval. This was done by using six polyethylene tanks containing rocks in a closed water circulating system. Two grams of Cladophora were introduced at 5:00 p.m. into each test tank containing four crayfish. At the end of the 4-and 16-hour periods the Cladophora filaments found in a form large enough to pick up with forceps were termed retrievable. Filaments that were too small to pick up and lying on the bottom were referred to as broken. The percentage weights of the original weight introduced were added to that which was retrieved in order to get a more accurate estimate of the food intake of the crayfish.

The periphyton plates were introduced at 5:00 p.m. and removed at 8:00 a.m. the following day. They were dried and weighed as set forth in the general methods.

Growth Studies

All of the crayfish were collected and weighed every ten days. Dry weights were obtained by drying at 60°C for 24 hours, and ash free dry weights by drying at 500°C for two hours. The calories were determined by the use of a Parr Bomb Calorimeter.

Molting Studies

The crayfish were observed daily for evidence of molting. Exoskeletons were removed if found before they had been eaten, and frozen for future analysis if desirable.

Molting was a very infrequent occurrence in the experiments. Therefore it was decided to try to force molting to see if the different diets had an effect upon the ability to molt. With the aid of Dr. E. M. Zipf, the eyestalks were carefully removed by the use of a scalpel. It is well known that removing the eyestalks will often force the crayfish to molt. Following the removal of the eyestalks the crayfish were immersed in chilled stream water in order to prevent bleeding. Twelve crayfish were used in this experiment, and after their eyes were removed they were placed back in their respective aquaria along with the normal crayfish. An effort was made to be sure that there were plenty of crevices among the rocks for protection of these eyeless forms from cannibalism. The longevity, weight change, and molting were followed until the crayfish died.

Feces Studies

The feces were obtained by isolating 5 to 10 crayfish in approximately 10-liter containers with aerated stream water for a period of 6 hours. The fecal material was collected and dried at 60°C for 24 hours and stored for later analyses. This method for obtaining feces was carried out at varying times during the course of the experiment. Microscopic observations were made of the feces. Those feces from the crayfish fed on periphyton contained a great many dead diatoms. The protoplasm had been removed. The feces of those fed on *Cladophora* contained both green filaments and those that had only the cell walls remaining. Attempts were made to determine the calories in each of the types of feces by the use of a microbomb calorimeter.

Respiration Studies

Respiration studies were carried out every ten days in Plexiglas respirometer chambers. The chambers were similar in design to those of Mac Intire, et al. (39). The whole unit was housed in a plywood water jacket. Ten crayfish were placed in each chamber, which was lined with clean rocks. The dissolved oxygen concentrations in the chambers were recorded in darkness without experimental animals in order to obtain any background error. It was realized that this method is only a rough means of determining respiration rates, as inevitably during the time in which the crayfish are in the tank bacterial growth occurs and error is introduced. However, since all the experiments were carried out in the same manner, the estimates are good for comparative purposes and believed to be fairly accurate.

Fish Experiments

3. Pimephales promelas Experiments

The diets of the fish in these experiments were unialgal *Cladophora* and *Cladophora* with epiphytes. The experiments were conducted in a controlled temperature room with a temperature between 20°-28°C, and a photoperiod of 12 hours.

A closed recirculating water system was built for each diet. This closed system consisted of two 200-liter glass aquaria, each with a water filter, attached in parallel to a 90-liter reservoir. One submersible pump at the bottom of each reservoir delivered the water to the aquaria at the rate of 2-3 liters/min/aquarium. The water was returned to the reservoir through a filter of loose cellulose fiber by gravity flow. Each week 30% of the water in each recirculating system was replaced by fresh stream water which was at the experimental temperature. One month after the start of the experiment the glass aquaria were replaced by 20-liter polyethylene tanks (18 cm x 45 cm x 23 cm). This was done to facilitate the collection of feces which was very difficult in the glass tanks. The replacement water and the original water in the unialgal Cladophora and the fish food diets were filtered through a 10-15 u filter in order to keep down contamination by other organisms.

In these experiments two size classes were used. The fish were sorted, and 20 fish of each size class were placed on each diet. Each fish was given an identifiable mark by injecting India ink from a 1 cc syringe with a 26 gauge needle into the caudal fin. The tattoos were performed while the fish was held on its side with light fine mesh nettings in approximately 0.5 cm water, which allowed the fish to continue breathing. The marks remained visible for approximately 3 weeks, whereupon they were remarked.

Food was introduced at 5:00 p.m. and removed at noon the next day. Wet-weighed amounts of periphyton and *Cladophora* with epiphytes were renewed daily (see general methods). The unialgal *Cladophora* was reused as so little was eaten, and replaced with new algae twice a week. Always more food was introduced than could be consumed and the remaining food was removed and weighed. Periphyton was introduced as growths on Plexiglas plates (13 cm x 23 cm x 2 mm) while the unialgal and *Cladophora* with epiphytes were anchored to the bottom by short glass rods.

Growth Studies

The fish were wet-weighed individually in a small transparent plastic bag partially filled with water, at periodic intervals during the experiment. Dry weights were obtained only on fish used to determine respiration rates. After the fish were killed in hot water they were dried at 60°C for

48 hours, and ash free dry weights were obtained by drying at 500°C for two hours. Individual and average wet weight values of fish are used to assess growth over time.

Respiration Studies

Respiration determinations were conducted on fish selected at random from each experimental diet. One-quart Mason jars served as the test chambers. The jar lids were modified to accommodate two 0.5 cm diameter glass tubes through which water was delivered to the bottom of the jar and exited at the top. The flow rate was regulated with a stopcock and set to deliver between 6 and 8 ml/minute/jar. The water was delivered by gravity directly to each jar through 0.5 cm diameter rubber tubing from a reservoir located above the test chambers. The flow rates remained relatively constant by maintaining a constant volume or head pressure in the reservoir. A submersible pump and several 100-l reservcirs at ground level supplied the head tank. Black plastic bags enveloped each jar in darkness to prevent algal growth.

Two fish were placed in each test chamber 72 hours prior to the test with test diet food. Twenty-four hours prior to the determinations all feces and remaining food was removed, and time was allowed for more than one complete replacement of water. The lids were examined for leaks and the desired experimental flow rates were set (6-8 ml/minute/jar). During the acclimation period, flow rates were 20-30 ml/minute/jar.

At the start of the test water samples (approximately 130-140 ml) were collected from the head tank and the water leaving the test chambers. Water was siphoned separately out of the head tank,and in all collections approximately 30-50% of the total sample volume was allowed to overflow, in an effort to minimize the error introduced by sample aeration. Given a constant known flow rate and knowing the oxygen concentration entering and leaving the test chamber, the oxygen consumption by the fish can be calculated. Oxygen determinations were performed using the standard Winkler method.

Well water was used during the respiration determinations to minimize the background respirations.

Caloric determinations were made by the use of a Parr Bomb Calorimeter (36) and a microbomb calorimeter (38).

^{14}C Uptake Studies

Diets were labeled by exposing Cladophora, Cladophora plus epiphytes, and periphyton (approximately 12.5 g/wet weight each) to 15 μ Ci $NaH^{14}CO_3$ for photosynthetic incorporation. After 4-hour incubation at 20°C, incorporated radioactivity was determined. Fifteen samples of each algal type were taken, dried, exposed to HCl fumes to remove sorbed ^{14}C, and weighed. The samples (approximately 5 mg dry weight) were combusted in a sample oxidizer (Packard Inst., Naperville, IL) and radioactivity was determined using a liquid scintillation counter. The bulk of the algae was transferred to fresh water and held overnight to permit release of labile excretion products and stabilize radioactivity levels. Having verified that the diets were sufficiently labeled for feeding studies from the initial check of radioactive content, the diets were again sampled and the radioactive content used as the initial level for each diet. Fish were fed as in feeding studies with unlabeled algae. Each day feces were collected, dried, weighed and oxidized as described for algae. Each day fish were transferred to clean water, killed, and blotted dry. The fish heads, guts, and viscera were dissected and the musculature was cut into several pieces of appropriate size for oxidation. All samples were dried, weighed, and oxidized for radioactive content as described. At the end of the experiment the diets were again sampled for radioactive content. The ash content of all materials was determined from weights after combustion at 550°C for four hours.

4. Ictalurus punctatus Experiments

Twenty fish were placed in each of six 55-gallon glass aquaria to acclimate for four days before the start of the experiment. Fish in two aquaria were placed on each of the following diets: periphyton and unialgal Cladophora. Fish in two aquaria were sacrificed for starting weights. Temperature and pH were recorded daily; oxygen and ammonia, one to three time a week; nitrate and nitrite, on two occasions.

From time to time microscopic observations were made of the periphyton on the plates that were supplied as food. The periphyton was dominated by Melosira varians with large populations of Navicula, Nitzschia, and Synedra, and smaller populations of Diatoma, Gomphonema, and other genera. Ciliated, flagellated, and amoeboid protozoans were represented in varying densities. Filamentous green algae such as Spirogyra and Stigeoclonium were occasionally observed and removed prior to introduction into the aquaria.

Fecal meterial was collected by isolating individuals in containers with filtered stream water for several hours. Feces were collected and microscopically observed for contents and algal condition.

Growth was determined by weighing the fish in tared plastic vessels containing a minimum of water on a triple beam balance. Wet weights were recorded to the nearest one-hundredth of a gram. All fish in each aquarium were removed and weighed at approximately one-week intervals.

5. Tadpole Experiments, *Rana pipiens*

These studies were carried out in polyethylene 20-liter trays. The diets consisted of cultured *Claodphora*, *Cladophora* plus epiphytes, and diatom-dominated periphyton. Determinations of pre-experiment weight and dry weights were carried out on a random sample of 40 tadpoles from an initial 280 individuals. Twenty-five tadpoles in each of two trays per diet were maintained.

During the first four weeks of the experiment water was metered in by a syringe pump at a rate of approximately 8 liters per hour. Overflow was returned to a 100-liter reservoir. Turnover time in the trays was approximately 2.25 hours. The closed system used during the last four weeks of the experiment was described in the methods for the snail experiments. Water in the trays was analyzed for temperature, NH_4, dissolved oxygen, and pH.

Periphyton plates were replaced daily. *Cladophora* was introduced daily in the *Cladophora*-plus-epiphytes diet and two to three times a week in the *Cladophora*-diet trays.

Every two weeks four or five tadpoles per tray were removed to determine wet and dry weights. Feces were collected daily for microscopic observations and later chemical analyses.

Two respiration measures are reported. A GRP 20 Gilson differential respirometer was used, and a temperature of 18°C was maintained during the test. Mean temperature in the experimental trays was 20°C.

RESULTS

All of these experiments were run in the natural soft waters of White Clay Creek, Pennsylvania. The oxygen was maintained above 7 ppm, the pH was circumneutral (7-8.2) and the ammonia was less than 40 µg/l.

Snail (*Physa heterostropha*) Studies

These studies to determine the suitability of *Cladophora glomerata* as a food for the snail *Physa heterostropha* were carried out over a three-year period, 1976-1978. The most extensive studies were made in 1978, when an attempt was made to relate the amount of food taken in and assimilated to that dissipated in respiration and excretion and utilized in growth. The experiments in 1976 were to determine the best design of experimentation, and were part of a series of preliminary studies on several organisms.

In 1978 four sets of experiments were performed: in one series the snails were fed on periphyton; in a second, the food was *Cladophora glomerata* plus epiphytes; in a third, unialgal *Cladophora glomerata* was the food; and in a fourth, the snails were starved. In the January 25-February 10 experiments the mean weight of the snails decreased in all experiments except those in which periphyton was the food (Tables 1, 2). The variance was large and this obscured the trend. The starved snails had an average decrease in mean ash free dry weight of 58.7% by the end of the experiment; the unialgal *Cladophora*-fed snails, 50.1%; and the snails with *Cladophora* plus epiphytes as a food source, 21%. The decrease in caloric value per snail showed a similar trend with the greatest loss in the starved and the unialgal *Cladophora*-fed snails. As seen in Tables 1 and 2, the calories per mg of tissue were quite similar on all diets, which indicates the correlation of increased caloric values with growth (Table 2).

A study of the shells showed a similar trend in that the increase in weight of the shell was greatest for the snails on the periphyton diet. On all diets the weight of the shell increased, but this increase was very little for the starved and *Cladophora*-fed snails.

A statistical analysis (40) of the dry weights of the snails on January 31 and February 10 showed the following relations of the tissue weights on the following diets: starved < *Cladophora* < *Cladophora* + epiphytes < periphyton (significant at $P < 0.001$). On January 31 lower weights of the starved snails ($P\ 0.05 > P > 0.01$) and higher weights of the periphyton-fed snails ($P < 0.001$) were significantly different from the pre-experiment weights. By February 10 the starved and *Cladophora*-fed snails were significantly lower ($P < 0.001$) in weight, the *Cladophora*-plus-epiphytes-fed snails were slightly but not significantly lower in weight, and the periphyton-fed snails continued to be significantly higher in weight than the pre-experiment snails.

The May 19 to July 14, 1977, experiment was much longer than the 1978 experiment (January 25 to February 10.) However, there were similar trends with a great increase in weight of tissue of those snails fed on periphyton and a decrease in tissue in those fed on unialgal *Cladophora*. Over a similar time span (14 days) the 1977 periphyton-fed snails increased 11.3 times in weight and 11.8 times in caloric value, as compared to 1978 with 2.1 times weight increase and 2 times increase in calories. This greater increase in 1977 was probably due to better environmental conditions for growth in May-July, and to the smaller size of the May-July snails. The caloric values over a similar time period averaged per mg ash free dry weight was 5.8 calories in 1977 and 5.24 calories in 1978 for the periphyton-fed snails, and 5.8 in 1977 and 5.63 in 1978 for the *Cladophora*-fed snails. There was not as much variability in the calories per mg of snail tissue as there was in the whole snail. As in the 1978 experiments, the value differences in the whole snail were due to growth (Tables 3, 4).

The loss in weight of the *Cladophora*-fed snails for the above time period was 15.1% in 1977 compared to 50.1% in 1978, and the decrease in caloric content of the snail tissue was 10% in 1977 as compared to 47.8% in 1978.

In 1976 similar experiments were performed in August-October. The size of the snails was intermediate between those used in 1977 and 1978. The percentage increase in dry weight over a similar period of time (20 days) was 5.6 times for the periphyton-fed snails and 5.8 times in caloric value. The *Cladophora* was not entirely free of epiphytes, but snails fed on this food produced similar results to those fed on unialgal *Cladophora* in 1978--that is, a loss of 42.4% in 1976 and 50.1% in 1978. The average calories lost per snail was 32% as compared to 47.8% in the 1978 *Cladophora* feeding experiments (Tables 5, 6).

Respiration Studies

For the 1978 experiments we have two sets of respiration studies--one set before the experiments started and one set on all diets during the experiment. An analysis of variance of the January 31 data shows the following ordering of differences among the groups ($P\ 0.05 > P\ 0.01$): starved < *Cladophora* < *Cladophora* plus epiphytes < periphyton (Table 7).

In the 1977 experiments we have four sets of data, all gathered during the experiment. Snails fed on *Cladophora* had significantly lower ($P < 0.05$), respiration rates than those fed on periphyton on June 2, 16, and 30 (Table 8).

An examination of the 1978 data shows an increase in respiration rates on all diets with the highest rate being on those snails fed on periphyton and the least increase in those snails that were being starved. In the 1977 and 1978 experiments the snails feeding on *Cladophora* had a lower respiration rate than those feeding on periphyton. The four sets of data obtained in 1977 are somewhat irregular. The rate of the periphyton-fed snails seemed to tend to decrease as the snails grew, whereas in the *Cladophora*-fed snails the trend was not as evident, as there was one very low rate on June 30 and a very high one on July 14. This may well be due to experimental error (Table 8).

No respiration studies were made during the 1976 experiments.

Food Intake

An effort was made to try to determine the food intake per day and the relative caloric value. It was realized that caloric value is only one measure of the nutritional value of food. For example, the caloric value of two very different carbohydrates might be very similar but the digestibility of the compounds very different.

One of the main problems was to determine the amount the snails actually ate. Wet weights of the food were carefully determined, but once it was presented to the snails considerable amounts may have been lost and not eaten. *Cladophora* often broke up into very short filaments which were practically uncollectable, and they were very difficult to see or to filter out of the feeding tanks. The diatoms were sometimes loosened in the feeding process and lost. Estimates of how much the snails ate each day are very rough.

The caloric determinations were made by the microbomb calorimeter technique. These are set forth in Tables 9 and 10. Our estimates of the caloric value of the diatoms are a little low as compared with some values in the literature. Our low values are probably due to fine silt which we could not separate from the diatom mat.

Two sets of data were obtained for *Cladophora* plus epiphytes and one set for unialgal *Cladophora*. As seen in Table 9, many periphyton values were determined. These data were only collected for 1978, but it is very probable that the caloric values for *Cladophora* (the same strain) and diatoms were very similar in 1977. As found by Paine and Vedes(5) the caloric values of diatoms and *Cladophora* are not very different.

Fecundity

In order to estimate the fecundity of the snail eggs and egg masses, they must be collected the day previous to the study period and, if possible, during the study, but always at the end of the period. When one examines Tables 1 and 11 and 3 and 12 it is evident that the difference in dry weight of tissue, egg production, and egg mass production are correlated.

In 1978 tissue weights only extend to February 10. Within that period the greatest increase in eggs/snail on the periphyton diet is correlated with the greatest increase in tissue weight (January 25 - 31, Tables 2, 11). However, the largest numbers of eggs per snail were in the periods February 12-14 and February 17. The weight increase was much less between January 31 and February 10. The egg masses per snail were largest on February 17, but the percentage increase was greatest between January 25 and January 31 (Table 11). On the *Cladophora* plus epiphytes diet the eggs per snail were much less,and usually less than one egg mass per snail was produced. The snails on the *Cladophora* diet and those that were starved produced very few eggs.

Viability tests were run on eggs collected on January 27, February 1, and February 6, 1978. In all cases only a few eggs did not hatch.

The caloric value of the eggs as determined from the 1978 experiments showed little variation on the different diets (Table 14).

In 1977 (Tables 3, 12) when one compares increased tissue weight with increased production on the periphyton diet, the greatest increase in tissue weight occurred between May 19 and June 2. This was also the period of greatest increase in eggs and egg masses. There was a steady increase in tissue weight per snail through July 14, but the increase between June 30 and July 14 was only 4 percent. The egg production per snail increased through June 30 and showed a 14% decrease on July 14. The production of egg masses per snail did not show such a consistent pattern of increase until June 30, but did show a decrease toward the end of the experiment.

In contrast, the snails fed unialgal Cladophora showed a decrease in weight and very few eggs and egg mass production as of June 2, and no eggs were produced after that date except for eight eggs on July 13.

The 1976 experiments (Table 13) showed the same general pattern of the 1977 and 1978 tests. The periphyton-fed snails increased in tissue weight, eggs, and egg masses, whereas the snails on the Cladophora plus epiphytes laid a few egg masses irregularly for a few days, and no eggs were laid after September 8, the 14th day of the experiment. The tissue dry weight of the snails steadily decreased.

Feces

In Table 15 is given the dates on which feces were collected in the 1977 and 1978 experiments. An inspection of this table shows that the feces of periphyton-fed snails had a mean ash free dry weight which was 84.5% (1978) to 86.2% (1977) of the dry weight. For the unialgal Cladophora-fed snails the mean percent varied from 54.4 (1977) to 60.6% (1978). The snails fed on Cladophora plus epiphytes had a mean percentage ash free dry weight of 73.7%, and for the starved snails 73.5% of the dry weight. These data indicate that the greatest amount of organic matter was in the feces of the periphyton-fed snails and the least in those snails fed on unialgal Cladophora.

Because there was very little organic matter in the feces, we were not able to determine the caloric content using the microbomb calorimeter.

Utilization of Calories

An attempt has been made to determine how the calories assimilated were partitioned in the 1978 and 1977 experiments (Tables 16, 17).

In 1978 the total calories assimilated by snails on the periphyton diet between January 25 and 31 were 52.35 cal/snail. Of this amount 27.7% was weight gained, 28.3% respired, and 44% was used in egg production. Only 17.6% of the food ingested on a calorie basis was assimilated (Table 16). On the Cladophora plus epiphytes diet, the amount of calories ingested was only 21% of those ingested on the periphyton diet. The snails lost weight. If we add calories in lost weight to those utilized in respiration and egg production, the partitioning is 33.3% for weight loss, 41.4% for respiration, and 25.3% for egg production. On the Cladophora diet and for those snails that were starved the calories used in respiration and egg production were less than in the periphyton and Cladophora plus epiphytes diets.

In the 1977 experiments we can only examine the partitioning of assimilated calories into growth or loss of weight and respiration (Table 17). On the periphyton diet the snails increased in weight throughout the experiment, but the calories partitioned to weight gain decreased from June 2, and those used in respiration increased. On the *Cladophora* diet the calories assimilated were much less. There was a decrease in caloric value of the snail tissue except in the period of June 30 to July 14, when there was a slight gain in weight from the June 30 weight. The percent of calories utilized in respiration showed an irregular trend, but were generally high which perhaps indicates that the snails were stressed by this diet. The data for June 16-30 is quite anomalous to the findings on the other dates.

Crayfish (Orconectes propinquus) Studies

The results of the crayfish experiments showed from a study of the mean weights that the crayfish fed on periphyton gained weight whereas those fed on *Cladophora* plus epiphytes and on unialgal *Cladophora* lost weight. However, in all cases the standard deviation was large enough to mask any real trend (Tables 18 and 19). The males tended to be heavier throughout the experiment. Very little molting occurred. It was quite evident that these three diets were not satisfactory and that the periphyton was a little better diet than the other two.

Respiration rates on all three diets declined during the course of the experiments (Table 20). This may have been at least in part due to the fact that the crayfish had recently molted when the experiment started and they molted very little during the course of the experiments. It is also known that crayfish often go into hiberation during the cooler months. The respiration rates were always higher in both males and females fed on periphyton. Respiration was a much larger percent of the food ingested on *Cladophora* and *Cladophora* plus epiphytes than it was for those crayfish fed on periphyton (Table 26).

The food consumed on a caloric basis was much greater on the periphyton diet than on the other two diets (Table 21, 22). On all diets the amount consumed decreased over the experiment. The females tended to ingest more food than the males, although this was not always true. It is very difficult to obtain an accurate estimate of the amount of food ingested due to breakage. An experiment was carried out to determine just how much this breakage was (Table 23). It was apparent that the amount of breakage was very variable, being 1-21% of the food presented.

Studies were made of the dry weight of the feces collected on any one day (Table 24). It was evident that the feces per crayfish was greater from those crayfish fed on periphyton. This one would expect, because of the much larger amount of food ingested. The average amount of feces produced from the Cladophora plus epiphytes diet and the Cladophora diet were not significantly different because of the very large variance which occurred. One can only say from these studies that the mean weights of the feces were less in these diets than in the periphyton diet; however, the large variance may negate this statement.

A study was made to determine how the calories ingested were partitioned. A much larger percentage of the food ingested seemed to have been assimilated on the Cladophora plus epiphytes and the Cladophora diet than in the experiments where the crayfish were fed periphyton (Table 25). The only data we have for all three diets is on October 3-4. The male and female crayfish lost weight on the Cladophora plus epiphytes diet but not on Cladophora. It is probable that on the Cladophora plus epiphytes diet they assimilated most of the food they ingested. On the Cladophora diet the females assimilated only 67.5% of the food ingested, whereas the males assimilated most of the food ingested.

On October 3 and 4 the crayfish on the three diets respired a similar percentage of the calories assimilated, but on December 13 and 14 the percentage of calories assimilated which were respired was less on the Cladophora plus epiphytes diet than on the other two diets.

At the end of the experiment the X organ-sinus gland of the eyestalk was removed to see if we could force molting. Molting did occur in the females on the periphyton diet (Table 26), and there was a gain in weight of the crayfish. It was difficult to determine whether the crayfish did not molt because of the period in their life cycle when the experiments were run or whether it was because of unsatisfactory conditions in the test experiments. Both factors might have been involved. However, it is evident from these experiments that the periphyton was a better diet for the crayfish than was the Cladophora or the Cladophora with epiphytes. If one bases his conclusions on weight, it was quite evident that they ingested more food on the periphyton diet than on the Cladophora plus epiphytes, and the latter was the better diet than Cladophora alone.

Tadpole (Rana pipiens) Studies

The results of the feeding experiments with the tadpoles Rana pipiens are set forth in Table 27. It is evident from Table 27 that those tadpoles fed on diatom-dominated periphyton grew much faster than those on Cladophora. The periphyton-fed tadpoles continued to increase rapidly in weight throughout the experiment. The tadpoles fed on Cladophora plus epiphytes grew at about the same rate as those fed on diatom-dominated periphyton alone. There is a little evidence that they even grew somewhat better when one compares the actual weights; however, the standard deviation is so great that these differences are really not significant. In contrast, the tadpoles fed Cladophora decreased from their starting weights until May 18 when they grew between May 6 and 18 to be at about the same weight as they were at the start of the experiment.

The tadpoles fed on periphyton showed signs of metamorphosing toward the end of the experiment. This was also true of those fed on Cladophora plus epiphytes. Those fed on Cladophora showed no signs of metamorphosing.

It was also noted that the tadpoles feeding on the Cladophora with epiphytes were just feeding on the epiphytes, as an examination of their intestines showed that diatoms were the main food passing through the gut. Because the diatoms were rather plentiful on the Cladophora and the tadpoles have the ability to scrape the diatoms from the Cladophora, it is understandable that these growth rates should not be different.

An examination of their respiration rates (Table 28) showed that the tadpoles fed on diatom periphyton has a similar respiration rate to those fed on Cladophora plus epiphytes, and these rates were lower than those fed on Cladophora alone. Those tadpoles fed on periphyton and those fed on Cladophora plus epiphytes had a decrease in respiration rate between April 5 and 18, whereas those fed on Cladophora alone had an increase in respiration rate. When we look at the calories consumed per tadpole in respiration those fed on Cladophora having very little weight consumed the greatest amount of oxygen. These small tadpoles which were fed Cladophora had a much higher respiration rate than those which were on a satisfactory diet, which may be correlated with the fact that they were under stress due to starvation.

Catfish (Ictalurus punctatus) Studies

These experiments were started on June 17 and terminated on July 15. They were cursory experiments simply to determine whether Ictalurus punctatus would eat Cladophora.

The experiments (Table 29) showed that the catfish on the periphyton diet had gained weight on July 1, continued to gain weight on July 8, but lost weight on July 15, so that their weight was similar to that at the start of the experiment.

On the Cladophora diet there were only two weighings after the start of the experiment, because there was a steady decrease in the size of the fish and they were about half their weight at the termination on July 8 than they were at the beginning of the experiment.

Fathead minnow (Pimephales promelas) Studies

The results of the experiments (March 7-May 23, 1978) in which periphyton, Cladophora and epiphytes, and unialgal Cladophora were fed to the fathead minnow, *Pimephales promelas*, produced several interesting results as to growth, assimilation of food ingested, and respiration.

In Tables 30 and 31 the growth of the fish throughout the experiment is given. The variance of the mean is fairly large, but it is clear that the fish on the periphyton diet grew throughout the experiment. Those fish on unialgal *Cladophora* or on *Cladophora* plus epiphytes lost weight throughout the experiment with the greatest loss in terms of calories between April 14 and May 23. This difference in growth is probably due to the difference in food consumed which was measured by food loss between the food presented and the food remaining after feeding (Tables 33, 34), and by experiments using ^{14}C-labeled food (Table 37). The average calories of food ingested in the two trays on the periphyton diet was approximately 23 times that ingested on the *Cladophora* plus epiphytes diet and 227 times the calories ingested on unialgal *Cladophora*.

As seen from Table 35, the average assimilated calories of periphyton as estimated from gain in weight and respiration was 6.2% of the food ingested March 7 to April 14, and 6.8% of ingested food from April 14 to May 23. This was determined by estimating the food consumed on a daily basis from Table 33 and the food assimilated for the number of days in the time interval from data in Table 35. In the *Cladophora* plus epiphytes diet these percentages were 221% by April 14 and 246% for April 14-May 23. For the *Cladophora* diet the calories assimilated of those ingested averaged 1717% as of April 14 and 2079% between April 14 and May 13, when all fish died. This is to a large measure the cause of the great weight losses on the two *Cladophora* diets. These data can only be considered as approximate, for although several attempts were made to determine respiration, reliable data were only obtained for one date (Table 32).

The assimilation of food as determined by experiments with ^{14}C (Tables 36, 37) show data which support the above relationship between food assimilated on the various diets. The ^{14}C uptake of the tagged food was less on an ash free dry weight (AFDW) basis than for the two *Cladophora* diets. The assimilation ratio--i.e., counts in muscles / counts in guts--was 0.14 for *Cladophora* and epiphytes versus 0.83 for periphyton (May 25) and 0.19 versus 0.42 (May 26) (Table 36). The intake of food (DPM/mg AFDW) on unialgal *Cladophora* was much less and so variable that not much significance can be attached to the data. In Table 37 an attempt is made to show the distribution of the food consumed in the fish. Generally the largest DPM/mg is in the muscle tissue and the least amount is in the viscera. It is interesting that the feces have a higher count in the *Cladophora* plus epiphytes diet than in the periphyton diet, and would indicate that of the food consumed much less was digested. As one would expect (although the data are not very reliable) much less food is ingested in the *Cladophora* diet.

The respiration rates from the data in Table 32 are least for fish on the unialgal *Cladophora* diet and greatest for those on the *Cladophora* plus epiphytes diet. The increase in weight (calories) on the periphyton diet as a percent of the calories respired varied between 25.8-30.6% (Table 35). The loss in weight (calories) as a percent of the calories respired on the *Cladophora* plus epiphytes diet was 34.1-72%, and on the unialgal *Cladophora* diet, 58-102.5%

The results of these experiments clearly showed that unialgal *Cladophora* and *Cladophora* plus epiphytes were unsatisfactory diets for the fathead minnow, *Pimephales promelas*. These data plus those on snails showed the importance of the chemical analyses of the food values of C. glomerata and the identifications, if they existed, of the presence of toxic chemicals. The studies of Larson (Part II) elucidate these concerns.

TABLE 1. DATA ON SNAIL TISSUE, 1978 EXPERIMENTS

	Dry Weights mg/snail		Ash Free[a] Dry Weight mg/snail		Ash Free[b] Dry Weight cal/snail	
Start of Experiment Jan. 25	3.484	4.434	3.205	4.079	17.256	21.961
	6.214	2.122	5.717	1.952	30.780	10.510
	4.270	4.852	3.928	4.464	21.148	24.034
	3.176	7.276	2.922	6.694	15.732	36.040
	3.490	3.408	3.211	3.135	17.288	16.879
	4.966	4.156	4.569	3.824	24.599	20.588
	5.114	5.068	4.705	4.663	25.332	25.106
	10.216	2.992	9.399	2.753	50.604	14.822
	4.240	4.572	3.901	4.206	21.003	22.645
	6.216	4.788	5.719	4.405	30.791	23.717
	5.412	3.052	4.979	2.808	26.807	15.118
	6.120	3.626	5.630	3.336	30.312	17.961
	4.398	4.084	4.046	3.757	21.784	20.228
	3.328	3.840	3.062	3.533	16.486	19.022
	7.160	6.552	6.587	6.028	35.464	32.455
	5.824	5.176	5.358	4.762	28.847	25.639
$\overline{X}$	4.800		4.417		23.78	
SD	1.61		1.47		7.93	
N	32		32		32	

[a] AFDW based on 92.0% of dry weight, see Table 2.
[b] 5.3841 cal/mg AFDW, see Table 2.

Periphyton Jan. 31	12.108		10.982[c]		52.725[d]	
	11.148		10.111		48.544	
	8.800		7.982		38.322	
	10.364		9.400		45.130	
	8.856		8.032		38.562	
	4.560		4.136		19.857	
	7.816		7.089		34.035	
	6.648		6.030		28.950	
$\overline{X}$	8.790		7.970		38.258	
SD	2.46		2.234		10.801	
N	8		8		8	

[c] AFDW based on 90.7% of dry weight, see Table 2.
[d] 4.8011 cal/mg AFDW, see Table 2.

(continued)

TABLE 1, continued.

	Dry Weight mg/snail	Ash Free Dry Weight mg/snail [e]	Ash Free Dry Weight cal/snail [f]
Periphyton Feb. 10	9.728	8.882	46.579
	6.020	5.496	28.822
	10.280	9.386	49.222
	7.338	6.700	35.136
	11.652	10.638	55.788
	10.240	9.349	49.028
	21.688	19.801	103.840
	4.336	3.959	20.762
$\overline{X}$	10.160	9.276	48.647
SD	5.269	4.811	25.258
N	8	8	8

[e] AFDW based on 91.3% of dry weight, see Table 2.
[f] 5.2442 cal/mg AFDW, see Table 2.

Cladophora + epiphytes Jan 31	4.100	3.485 [g]	18.701 [h]
	4.628	3.934	21.110
	3.252	2.764	14.832
	3.592	3.053	16.382
	7.068	6.008	32.239
	3.696	3.142	16.860
	4.264	3.624	19.446
	3.600	3.060	16.420
$\overline{X}$	4.280	3.634	19.499
SD	1.21	1.029	5.522
N	8	8	8

[g] AFDW values based on 85.0% of dry weight, see Table 2.
[h] 5.3664 cal/mg AFDW, see Table 2.

Feb. 10	4.400	4.083 [i]	21.617 [j]
	5.112	4.744	25.117
	2.528	2.346	12.421
	4.704	4.365	23.110
	3.908	3.627	19.203
	1.792	1.663	8.805
$\overline{X}$	3.741	3.471	18.379
SD	1.307	1.213	6.420
N	6	6	6

[i] AFDW based on 92.8% of dry weight, see Table 2.
[j] 5.2944 cal/mg AFDW, see Table 2.

(continued)

TABLE 1, continued.

	Dry Weight mg/l snail	Ash Free[k] Dry Weight mg/l snail	Ash Free[l] Dry Weight cal/snail
Cladophora Jan. 31	2.346	2.172[1]	10.431[2]
	4.830	4.473	21.481
	4.438	4.110	19.738
	2.782	2.576	12.371
	5.004	4.634	22.254
	3.590	3.324	15.963
	3.146	2.913	13.989
	4.198	3.887	18.667
$\overline{X}$	3.792	3.513	16.862
SD	0.978	0.904	4.351
N	8	8	8

[k] AFDW values based on 92.6% of dry weight, see Table 2.
[l] 4.8024 cal/mg AFDW, see Table 2.

Feb. 9	1.400	1.295[m]	7.290[n]
	2.376	2.198	12.373
	2.706	2.503	14.090
	3.000	2.775	15.621
	2.636	2.438	13.724
	2.184	2.020	11.371
$\overline{X}$	2.384	2.205	12.412
SD	0.558	0.516	2.904
N	6	6	6

[m] AFDW values based on 92.5% of dry weight, see Table 2.
[n] 5.6292 cal/mg AFDW, see Table 2.

(continued)

TABLE 1, continued.

	Dry Weight mg/l snail	Ash Free [o] Dry Weight mg/l snail	Ash Free [p] Dry Weight cal/snail
Starved (Day 6) Jan. 31	2.482	2.336	11.809
	2.146	2.019	10.206
	5.304	4.991	25.231
	2.280	2.145	10.843
	2.940	2.767	13.988
	3.472	3.267	16.515
	4.370	4.112	20.787
	2.062	1.940	9.807
X̄	3.130	2.947	14.898
SD	1.180	1.106	5.591
N	8	8	8

[o] AFDW values based on 94.1% of dry weight, see Table 2.
[p] 5.0552 cal/mg AFDW, see Table 2.

	Dry Weight mg/l snail	Ash Free Dry Weight mg/l snail	Ash Free Dry Weight cal/snail
(Day 15) Feb. 9	1.792	1.636 [q]	8.780 [r]
	2.406	2.197	11.791
	2.884	2.633	14.131
	0.930	0.849	4.557
	1.370	1.251	6.714
	2.592	2.366	12.698
X̄	1.996	1.822	9.779
SD	0.759	0.693	3.719
N	6	6	6

[q] AFDW values based on 91.3% of dry weight, see Table 2.
[r] 5.3671 cal/mg AFDW, see Table 2.

TABLE 2. DATA ON WHICH CALORIE CALCULATIONS ARE BASED, 1978 SNAIL TISSUE EXPERIMENTS

	Dry Weight*	Ash Free Dry Weight	Percent Ash Free Dry Weight	Ash Free Dry Weight cal/mg
Start of Experiment				
Jan. 24	9.30	8.76	94.2	5.1183
	6.48	5.94	91.7	5.2559
	10.52	9.68	92.0	5.2581
	7.67	7.19	93.7	5.3631
	8.34	7.36	88.2	5.7003
	8.58	7.91	92.2	5.6089
$\overline{X}$			92.0	5.3841
SD			2.11	0.2253
N			6	6
Periphyton				
Jan. 31	7.76	6.88	88.7	4.3416
	12.26	11.02	89.9	5.0948
	10.32	9.64	93.4	4.9668
$\overline{X}$			90.7	4.8011
SD			2.44	0.4030
N			3	3
Feb. 9	9.12	8.26	90.6	5.6160
	10.74	10.08	93.9	5.3502
	9.08	8.12	89.4	4.7664
$\overline{X}$			91.3	5.2442
SD			2.33	0.4346
N			3	3
Cladophora + epiphytes				
Jan. 31	7.72	7.09	91.8	5.0661
	10.80	9.22	85.4	4.8669
	8.00	6.22	77.7	6.1662
$\overline{X}$			85.0	5.3664
SD			7.1	0.6997
N			3	3
Feb. 9	6.36	5.90	92.8	5.4955
	8.56	7.98	93.2	5.0390
	8.28	7.64	92.3	5.3487
$\overline{X}$			92.8	5.2944
SD			0.45	0.2330
N			3	3

(continued)

TABLE 2, continued.

	Dry Weight	Ash Free Dry Weight	Percent Ash Free Dry Weight	Ash Free Dry Weight cal/mg
Cladophora				
Jan. 31	7.00	6.28	89.7	4.4470
	8.24	7.80	94.7	4.6904
	6.74	6.30	93.5	5.2699
$\overline{X}$			92.6	4.8024
SD			2.61	0.4227
N			3	3
Feb. 9	6.53	6.03	92.3	5.4796
	7.10	6.58	92.7	5.7788
$\overline{X}$			92.5	5.6292
SD			0.28	0.2115
N			2	2
Starved				
Jan. 31	6.68	6.42	96.1	5.2041
	6.46	6.04	93.5	4.7585
	6.34	5.88	92.7	5.2029
$\overline{X}$			94.1	5.0552
SD			1.78	0.2569
N			3	3
Feb. 9	6.18	5.74	92.9	5.6544
	4.68	4.20	89.7	5.0798
$\overline{X}$			91.3	5.3671
SD			2.26	0.4063
N			2	2

* Number of snails not known.

TABLE 3. DATA ON SNAIL TISSUE, 1977 EXPERIMENTS

	Dry Weight mg/snail		Ash Free[a] Dry Weight mg/snail		Ash Free[b] Dry Weight cal/snail	
Start of Experiment May 19	1.919	1.626	1.764	1.494	9.649	8.166
	0.168	0.788	0.154	0.724	0.842	3.957
	1.362	0.512	1.252	0.471	6.848	2.574
	0.204	0.588	0.187	0.540	1.023	2.952
	1.490	1.460	1.369	1.342	7.488	7.335
	0.422	1.818	0.388	1.671	2.121	9.134
	1.328	0.656	1.220	0.603	6.669	3.296
	0.808	0.758	0.743	0.697	4.061	3.810
	0.440	1.072	0.404	0.985	2.208	5.384
	0.208	0.672	0.191	0.618	1.044	3.378
	0.806	0.278	0.741	0.255	4.050	1.394
	0.718	0.600	0.660	0.551	3.608	3.012
	0.466	0.700	0.428	0.643	2.339	3.515
$\overline{X}$	0.840		0.773		4.225	
SD	0.510		0.468		2.562	
N	26		26		26	
Periphyton June 2	6.688	8.996	5.959	8.015[c]	34.373	46.233[d]
	5.364	15.353	4.779	13.680	27.567	78.911
	6.876	11.360	6.127	10.122	35.343	58.387
	11.800	10.424	10.514	9.288	60.648	53.576
	8.916	13.328	7.944	11.875	45.824	68.499
	12.096	8.648	10.778	7.705	12.171	44.445
	10.616	9.136	9.459	8.140	54.563	46.954
	7.324		6.526		37.644	
$\overline{X}$	9.79		8.727		50.041	
SD	2.70		2.420		14.437	
N	15		15		15	

[a] AFDW values based on 91.9% of dry weight, see Table 4.

[b] 5.4660 cal/mg AFDW, see Table 4.

[c] AFDW based on 92.8% of dry weight, see Table 4.

[d] 5.8082 cal/mg AFDW, see Table 4.

TABLE 3, continued.

	Dry Weight mg/snail		Ash Free[e] Dry Weight mg/snail		Ash Free[f] Dry Weight cal/snail	
June 16	14.316	17.248	13.371	16.110	68.851	82.955
	13.392	20.600	12.508	19.240	64.407	99.073
	18.856	17.336	17.612	16.192	90.689	83.377
	11.712	10.620	10.939	9.919	56.328	51.076
	12.396	22.032	11.578	20.578	59.619	105.960
	13.608	14.700	12.710	13.730	65.448	70.700
	6.832	15.584	6.381	14.555	32.858	74.948
	15.696		14.660		75.489	
$\overline{X}$	15.0		14.006		72.113	
SD	3.92		3.665		18.870	
N	15		15		15	
	[e] AFDW based on 93.4% of dry weight, see Table 4. [f] 5.1493 cal/mg AFDW, see Table 4.					
June 30	21.850	26.500	20.124	24.407[g]	99.854	121.110[h]
	32.800	26.650	30.209	24.545	149.900	121.790
	29.800	29.000	27.446	26.709	136.190	132.530
	20.300	24.400	18.696	22.472	92.769	111.500
	32.050	22.500	29.518	20.723	146.470	102.830
	44.500	15.000	40.985	13.815	203.370	68.549
	30.200	21.600	27.814	19.894	138.010	98.713
	23.000		21.183		105.11	
$\overline{X}$	26.68		24.569		121.91	
SD	6.94		6.390		31.71	
N	15		15		15	
	[g] AFDW based on 92.1% of dry weight, see Table 4. [h] 4.9620 cal/mg AFDW, see Table 4.					
July 14	53.100	47.400	46.728	41.712[i]	260.320	232.380[j]
	13.000	6.636	11.440	5.840	63.730	32.535
	34.792	6.142	30.617	5.405	170.570	30.111
	26.976	51.650	23.739	45.452	132.250	253.210
	22.192	16.052	19.529	14.126	108.800	78.696
	20.400	32.904	17.952	28.956	100.010	161.310
	37.400	41.000	32.912	36.080	183.350	201.000
$\overline{X}$	27.97		25.749		143.45	
SD	14.34		13.947		77.70	
N	14		14		14	
	[i] AFDW based on 88.0% of dry weight, see Table 4. [j] 5.5707 cal/mg AFDW, see Table 4.					

(continued)

TABLE 3, continued.

	Dry Weight mg/snail		Ash Free Dry Weight mg/snail [k]		Ash Free Dry Weight cal/snail [l]	
Cladophora June 2	0.468	0.338	0.434	0.314	2.521	1.824
	0.708	0.572	0.657	0.531	3.816	3.084
	1.098	1.034	1.019	0.960	5.918	5.576
	0.572	0.802	0.531	0.744	3.084	4.321
	0.700	0.658	0.650	0.611	3.775	3.549
	0.292	0.718	0.271	0.666	1.574	3.868
	0.964	0.928	0.895	0.861	5.198	5.001
	0.754		0.700		4.066	
$\overline{X}$	0.707		0.656		3.812	
SD	0.240		0.221		1.281	
N	15		15		15	
	[k] AFDW based on 92.8% of dry weight, see Table 4.					
	[l] 5.8082 cal/mg AFDW, see Table 4.					
June 16	0.898	0.788	0.823	0.722[m]	4.654	4.084[n]
	0.844	0.426	0.773	0.390	4.372	2.206
	0.642	0.372	0.588	0.341	3.326	1.929
	0.934	0.544	0.856	0.498	4.842	2.817
	0.346	0.418	0.317	0.383	1.793	2.166
	0.274	0.832	0.251	0.762	1.420	4.310
	0.746	0.256	0.683	0.234	3.863	1.323
$\overline{X}$	0.594		0.544		3.079	
SD	0.240		0.224		1.269	
N	14		14		14	
	[m] AFDW based on 91.6% of dry weight, see Table 4.					
	[n] 5.6556 cal/mg AFDW, see Table 4.					
June 30	0.250	0.300	0.231	0.278[o]	1.239	1.491[p]
	0.550	0.600	0.509	0.555	2.731	2.978
	0.650	0.400	0.601	0.370	3.224	1.985
	0.400	0.350	0.370	0.324	1.985	1.738
	0.470	0.100	0.435	0.093	2.334	0.499
	0.700	0.250	0.648	0.231	3.477	1.239
	0.150	0.350	0.139	0.324	0.746	1.738
	0.400		0.350		1.878	
$\overline{X}$	0.40		0.364		1.952	
SD	0.18		0.162		0.872	
N	15		15		15	
	[o] AFDW based on 92.5% of dry weight, see Table 4.					
	[p] 5.3651 cal/mg AFDW, see Table 4.					

(continued)

TABLE 3, continued.

	Dry Weight mg/snail		Ash Free[q] Dry Weight mg/snail		Ash Free[r] Dry Weight cal/snail	
July 14	0.474	0.762	0.416	0.669	2.033	3.270
	0.400	0.368	0.351	0.323	1.716	1.579
	0.488	0.804	0.428	0.706	2.092	3.451
	0.340	0.264	0.299	0.232	1.462	1.134
	0.184	1.000	0.162	0.878	0.792	4.292
	0.480	0.264	0.421	0.232	2.058	1.134
	0.348	0.524	0.306	0.460	1.496	2.248
$\bar{X}$	0.480		0.418		2.054	
SD	0.23		0.204		0.991	
N	14		14		14	

[q] AFDW based on 87.8% of dry weight, see Table 4.

[r] 4.8877 cal/mg AFDW, see Table 4.

TABLE 4. DATA ON WHICH CALORIE CALCULATIONS ARE BASED, 1977 SNAIL TISSUE EXPERIMENTS

	Dry Weight*	Ash Free Dry Weight	Percent Ash Free Dry Weight	Ash Free Dry Weight cal/mg
Start of Experiment				
May 19	7.16	6.58	91.9	5.6703
	5.88	5.40	91.8	5.2616
$\overline{X}$			91.9	5.4660
SD			0.07	0.29
N			2	2
Periphyton				
June 2	8.53	7.98	93.6	6.0491
	10.13	8.69	85.8	6.0332
	11.68	10.26	87.8	5.2227
$\overline{X}$			89.1	5.7683
SD			4.1	0.47
N			3	3
June 16	18.24	17.26	94.6	5.0270
	13.34	12.08	90.6	5.0209
	10.86	10.30	94.9	5.4000
$\overline{X}$			93.4	5.1493
SD			2.4	0.22
N			3	3
June 30	12.22	10.00	81.8	5.1832
	25.06	24.18	96.5	4.6084
	17.44	16.46	94.4	4.9204
	8.26	7.90	95.6	5.1358
$\overline{X}$			92.1	4.9620
SD			6.9	0.26
N			4	4
July 14	20.12	19.14	88.0	5.0179
	12.34	10.86	88.0	5.4432
	19.42	17.10	88.1	6.2510
$\overline{X}$			88.0	5.5707
SD			0.06	0.63
N			3	3

(continued)

TABLE 4, continued.

Cladophora	Dry Weight	Ash free Dry Weight	Percent Ash Free Dry Weight	Ash Free Dry Weight cal/mg
June 2	3.38	3.12	92.3	5.4799
	4.12	3.84	93.2	6.1365
$\overline{X}$			92.8	5.8082
SD			0.64	0.46
N			2	2
June 16	6.18	5.66	91.6	5.6556
June 30	4.82	4.46	92.5	5.3651
July 14	6.40	5.62	87.8	4.8877

* Number of snails not known.

TABLE 5. DATA ON SNAIL TISSUE, 1976 EXPERIMENTS

	Dry Weights mg/snail		Ash Free [a] Dry Weight mg/snail		Ash Free [b] Dry Weight cal/snail	
Start of Experiment August 24	1.476	1.105	1.312	0.982	5.865	4.390
	2.070	2.067	1.840	1.838	8.225	8.216
	3.000	2.412	2.667	2.144	11.921	9.584
	2.686	1.188	2.388	1.056	10.674	4.720
	1.680	2.891	1.494	2.570	6.678	11.488
	2.188	1.851	1.945	1.646	8.694	7.358
	1.607	2.447	1.429	2.175	6.388	9.722
	2.559	3.522	2.275	3.131	10.169	14.000
	2.969	2.536	2.629	2.255	11.796	10.080
	1.423	1.251	1.265	1.112	5.655	4.971
	2.973	1.832	2.643	1.629	11.814	7.282
	1.157	0.818	1.029	0.727	4.600	3.250
$\overline{X}$	2.07		1.841		8.231	
SD	0.73		0.653		2.918	
N	24		24		24	

[a] Actual dry weights and wet weights of snails.
[b] 4.4742 cal/mg AFDW, see Table 6.

	Dry Weights mg/snail	Ash Free Dry Weight mg/snail	Ash Free Dry Weight cal/snail
Periphyton Sept. 3			
$\overline{X}$	6.23[c]	5.538[d]	24.778[d]
N	12	12	12
Sept. 13			
$\overline{X}$	11.42	10.358[d]	55.712[d]
N	12	12	12
Sept. 22			
$\overline{X}$	12.49	11.553[d]	57.640[d]
N	12	12	12
Oct. 5			
$\overline{X}$	18.20	14.724[d]	78.742[d]
N	12	12	12

[c] No individual weights for snails
[d] See Table 6 for values on which calculations are based.

(continued)

TABLE 5, continued

	Dry Weights mg/1 snail	Ash Free Dry Weight mg/1 snail	Ash Free Dry Weight cal/snail
Cladophora			
Sept. 3			
$\bar{X}$	1.46[e]	1.348	4.839
N	12	12	12
Sept. 13			
$\bar{X}$	1.30	1.060	5.58
N	12	12	12
Sept. 22			
$\bar{X}$	0.79	0.639	3.281
N	12	12	12
Oct. 5			
$\bar{X}$	0.81	0.710	3.76
N	12	12	12

[e] No individual weights for snails

TABLE 6. DATA ON WHICH CALORIE CALCULATIONS ARE BASED, 1976 SNAIL TISSUE EXPERIMENTS

	Dry Weight*	Ash Free Dry Weight	Percent Ash Free Dry Weight	Ash Free Dry Weight cal/mg
Start of Experiment				
Aug. 24		no data		
Periphyton				
Sept. 3	16.34	15.94	97.6	4.1248
	14.78	14.16	95.8	4.6326
	8.58	6.30	73.4	4.6651
$\overline{X}$			88.9	4.4742
SD			13.5	0.30
N			3	3
Sept. 13	11.08	9.98	90.1	5.3786
	7.78	7.10	91.3	5.1714
$\overline{X}$			90.7	5.2750
SD			0.9	0.15
N			2	2
Sept. 22	21.82	19.64	90.0	5.1124
	18.28	17.34	94.9	4.8659
$\overline{X}$			92.5	4.9892
SD			3.5	0.17
N			2	2
Oct. 5	12.88	10.02	77.8	4.9616
	11.28	9.46	83.9	5.7341
$\overline{X}$			80.6	5.3479
SD			4.3	0.55
N			2	2
Cladophora	9.38	8.96	95.5	3.4595
Sept. 3	8.02	7.48	93.3	3.8957
	6.98	6.15	88.1	3.4132
$\overline{X}$			92.3	3.5895
SD			3.8	0.27
N			3	3

(continued)

TABLE 6, continued.

	Dry Weight	Ash Free Dry Weight	Percent Ash Free Dry Weight	Ash Free Dry Weight cal/mg
Cladophora				
Sept. 13	6.28	4.80	76.4	5.0175
	4.92	4.26	86.6	5.5146
$\overline{X}$			81.5	5.2661
SD			7.2	0.35
N			2	2
Sept. 22	6.28	5.08	80.9	5.1354
Oct. 5	3.76	3.34	88.8	5.8183
	3.22	2.78	86.3	4.7908
$\overline{X}$			87.6	5.3046
SD			1.8	0.73
N			2	2

* Number of snails not known.

TABLE 7. SNAIL RESPIRATION, 1978 EXPERIMENTS

	Dry weight of snail tissue (mg)			ul O_2/mg snail/hr			cal/ mg snail/ hr *	cal/ snail/ hr	cal/ snail/ day
	$\overline{X}$	SD	N	$\overline{X}$	SD	N			
Jan. 24									
Start of Experiment	4.80	1.61	32	1.25	0.20	8	.0059	0.028	0.672
Jan. 31									
Periphyton	8.79	2.46	8	2.49	0.11	2	.0117	0.103	2.472
Cladophora + Epiphytes	4.28	1.21	8	1.84	0.64	2	.0086	0.037	0.888
Cladophora	3.79	0.98	8	1.65	0.11	2	.0078	0.029	0.696
Starved	3.13	1.18	8	1.51	0.22	2	.0071	0.022	0.528

* 1 ul O_2 = 0.00472 calories

TABLE 8. SNAIL RESPIRATION, 1977 EXPERIMENTS

	Dry weight of snail tissue (mg)			ul O_2/mg snail/hr			cal/mg snail/hr *	cal/ snail/hr	cal/ snail/day
	$\overline{X}$	SD	N	$\overline{X}$	SD	N			
Periphyton									
June 2	9.80	2.70	15	2.54	1.16	3	0.012	0.118	2.822
June 16	15.00	3.92	15	1.99	0.46	3	0.0094	0.141	3.384
June 30	26.68	6.94	15	2.27	0.25	3	0.011	0.293	7.044
July 14	29.30	15.85	14	2.09	0.36	3	0.0099	0.290	6.962
Cladophora									
June 2	0.71	0.24	15	1.29	0.07	3	0.0061	0.0040	0.096
June 16	0.59	0.24	14	1.28	0.54	3	0.0060	0.0035	0.084
June 30	0.40	0.17	15	0.67	0.24	3	0.0031	0.0010	0.024
July 14	0.48	0.23	14	4.13	1.27	3	0.0194	0.0930	0.232

* 1 ul O_2 = 0.00472 calories

TABLE 9. SNAIL FOOD CONSUMPTION, 1978 EXPERIMENTS

Periphyton	Wet Wt. mg consumed	Dry Wt.[a] mg consumed	mg Ash Free Dry Weight[b]	No. of Snails	Mg AFDW/ snail	Calories[c] consumed/ snail
Jan. 29 - Tray 5	4790	2031	280.3	20	14.014	55.333
Tray 6	5100	2162	298.4	20	14.921	58.906
Jan. 30 - Tray 5	3900	1654	228.2	20	11.410	45.046
Tray 6	4100	1738	239.9	19	12.626	49.849
Jan. 31 - Tray 5	3100	1314	181.4	16	11.337	42.205
Tray 6	800	339	46.81	14	3.344	12.447
$\overline{X}$					11.275	43.964
SD					4.136	16.641
N					6	6
Feb. 1 - Tray 5	3720	1577	220.8	15	14.719	54.795
Tray 6	1200	509	71.23	10	7.123	26.518
Feb. 3 - Tray 5	2700	1145	160.3	15	10.685	39.777
Tray 6	2300	975	136.5	10	13.653	50.827
Feb. 4 - Tray 5	2300	975	136.5	15	9.100	33.877
Tray 6	3100	1314	181.4	10	18.140	67.532
Feb. 5 - Tray 5	1900	806	112.8	15	7.519	27.991
Tray 6	2000	848	118.7	10	11.870	44.190
Feb. 6 - Tray 5	3100	1314	181.4	15	12.093	43.360
Tray 6	2500	1060	148.4	10	14.840	56.891
Feb. 8 - Tray 5	2900	1230	172.1	14	12.296	47.138
Tray 6	2100	890	124.7	9	13.851	53.098
Feb. 9 - Tray 5	1300	551	77.17	10	7.717	29.583
Tray 6	700	297	41.55	5	8.310	31.859
Feb. 10 - Tray 5	1400	594	83.10	9	9.234	35.399
Feb. 13 - Tray 5	900	382	53.42	8	6.678	25.601
Tray 6	1000	424	59.36	4	14.840	56.891
$\overline{X}$					11.333	42.843
SD					3.372	12.685
N					17	17

[a] Dry weight based on 42.4% of wet weight.

[b] Ash Free Dry Weight based on 13.4% of dry weight for Jan. 29-31; 14.0% of dry weight on Feb. 1.

[c] Jan. 29-31 = 3.9484 cal/mg AFDW; Feb. 1-5 = 3.7228 cal/mg AFDW; Feb. 6-10 = 3.8336 cal/mg AFDW, see Table 10.

(continued)

TABLE 9, continued.

	Wet Wt. mg consumed	Dry Wt.[d] mg consumed	mg[e] Ash Free Dry Weight	No. of Snails	mg AFDW/ snail	Calories[f] consumed snail
Cladophora + Epiphytes						
Jan. 29 - Tray 7	36.82	25.37	25.37	20	1.269	5.594
Jan. 31 - Tray 7	2.10	1.45	1.45	13	0.112	0.494
Tray 8	67.33	46.39	46.39	11	4.217	18.589
Feb. 1 - Tray 7	33.66	23.19	23.19	9	2.577	11.359
Tray 8	31.56	21.74	21.74	11	1.976	8.710
$\overline{X}$					2.03	8.949
SD					1.528	6.737
N					5	5

[d] Dry weight based on 10.52% of wet weight.
[e] AFDW based on 68.9% of dry weight, see Table 10.
[f] 4.408 cal/mg AFDW, see Table 10.

Cladophora						
Jan. 29 - Tray 4	18.40	15.49[g]	15.49[h]	19	0.815	3.230[i]

[g] Dry weight based on 18.4% of wet weight.
[h] AFDW based on 84.2% of dry weight, see Table 10.
[i] 3.9615 cal/mg AFDW, see Table 10.

TABLE 10. DATA FOR BASIS OF CONVERSIONS FROM DRY WEIGHT TO CALORIES, 1978 SNAIL FOOD EXPERIMENTS

	Dry Weight mg/l	Ash Free Dry Weight mg/l	% Ash Free Dry Weight	Ash Free Dry Weight cal/mg
Periphyton				
Jan. 30	943.9	129.4	13.7	3.6909
	522.9	67.7	12.9	4.2059
	196.3	26.8	13.7	
$\overline{X}$			13.4	3.9484
SD			0.46	0.364
N			3	2
Feb. 1	915.5	124.2	13.6	3.4861
	914.6	126.6	13.8	3.9595
	312.6	189.7	14.5	
$\overline{X}$			14.0	3.7228
SD			0.47	0.335
N			3	2
Feb. 10	727.9	96.0	13.2	4.0036
	624.1	85.2	13.7	3.1842
	361.5	54.5	15.1	4.0745
				4.0722
$\overline{X}$			14.0	3.8336
SD			0.98	0.434
N			3	4
Cladophora + Epiphytes				
Feb. 1979	12.72	9.64	75.8	4.480
	13.74	10.18	74.1	4.358
	17.94	10.48	58.4	4.167
	19.30	10.94	56.7	4.406
	12.80	10.40	81.3	4.308
	14.98	12.06	80.1	4.470
	20.28	13.14	64.8	4.379
	19.24	11.50	59.8	4.699
$\overline{X}$			68.9	4.408
SD			10.1	0.1536
N			8	8

(continued)

TABLE 10, continued.

	Dry Weight mg/l	Ash Free Dry Weight mg/l	% Ash Free Dry Weight	Ash Free Dry Weight cal/mg
Cladophora				
Jan. 30	428.1	354.1	82.7	4.0447
	385.8	321.4	83.3	4.1909
	311.9	270.3	86.7	3.6489
$\overline{X}$			84.2	3.9615
SD			2.16	0.280
N			3	3

TABLE 11. SNAIL FECUNDITY, 1978 EXPERIMENTS

	No. Days	No. Eggs	No. egg Masses	No. Snails	Eggs/ snail/day	Egg masses/ snail/day
Periphyton						
Jan. 24-27	4	998	76	40	6.24	0.48
Jan. 28-30	3	2832	138	39.67	23.80	1.16
Jan. 31-Feb. 1	2	1828	88	27.5	33.24	1.60
Feb. 2-3	2	1827	83	25	36.54	1.66
Feb. 12-14	3	6559	113	25	87.45	1.51
Feb. 17	1	1136	34	14	81.14	2.43
Cladophora + Epiphytes						
Jan. 31-Feb. 1	2	506	38	22	11.5	0.86
Feb. 2-3	2	250	25	20	6.25	0.63
Feb. 12-14	3	427	29	19	7.49	0.51
Feb. 17	1	125	6	5	25.0	1.20
Cladophora						
Jan. 31-Feb. 1	2	83	11	20.5	2.02	0.27
Feb. 12-14	3	47	5	9	1.74	0.19
Starved						
Jan. 31-Feb. 1	2	82	12	22.5	1.82	0.27
Feb. 12-14	3	26	6	13.67	0.63	0.15

TABLE 12. SNAIL FECUNDITY, 1977 EXPERIMENTS

Dates & Diets	No. Days	No. Eggs	No. Egg Masses	No. Snails	Eggs/ Snail/Day	Egg Masses/ Snail/Day
Periphyton						
May 23	1	129	11	81	1.59	0.14
May 24-27	4	3357	186	81	10.36	0.57
June 7-8	2	5941	79	59	50.35	0.67
June 9-10	2	6378	123	59	54.05	1.04
June 11-13	3	9769	179	58	56.14	1.03
June 14-15	2	6384	93	58	55.03	0.80
June 16-17	2	5720	84	42*	68.10	1.00
June 18-20	4	10369	152	41	63.23	0.93
June 22-24	3	11221	145	40	93.51	1.21
June 25-27	3	10834	145	40	90.28	1.21
June 28-July 1	4	15687	184	32	122.55	1.44
July 6-8	3	10442	100	20.33*	171.21	1.64
July 9-11	3	7421	83	20	123.68	1.38
July 12-13	2	5022	55	20	125.55	1.38
July 14	1	2103	19	20	105.15	0.95
Cladophora						
May 23	1	0	0		0	0
May 24-27	4	0	0	81	0	0
May 31	1	33	6	79	0.42	0.08
June 3	1	24	6	64	0.38	0.09
June 7-8	2	0	0	63	0	0
June 9-10	2	0	0	63	0	0
June 11-13	3	0	0	63, 62	0	0
June 14-15	2	0	0	61	0	0
June 16-17	2	0	0	45, 44	0	0
June 18-21	4	0	0	43, 42	0	0
June 22-24	3	0	0	42	0	0
June 25-27	3	0	0	41	0	0
June 28-July 1	4	0	0	41, 40, 25	0	0
July 6-8	3	0	0	22	0	0
July 9-11	3	0	0	22	0	0
July 13	1	8	2	19	0.42	0.11
July 14	1	0	0	5*	0	0

*Drop in snail numbers due to removal for respiration experiments.

TABLE 13. SNAIL FECUNDITY, 1976 EXPERIMENTS

	No. Days	No. Eggs	No. egg Masses	No. Snails	Eggs/ snail/day	Egg masses/ snail/day
Periphyton						
Aug. 25	1	312	16	158	1.97	0.10
Aug. 26	1	196	10	156	1.26	0.06
Aug. 27	1	580	29	155	3.74	0.19
Aug. 28	1	1336	53	152	8.79	0.35
Aug. 31	1	1113	26	150	7.42	0.17
Sept. 1	1	2870	70	149	19.26	0.47
Sept. 10	1	3478	72	115	30.24	0.63
Sept. 14	1	1894	34	81*	23.38	0.42
Sept. 15	1	3526	64	81	43.53	0.79
Sept. 16	1	3400	65	80	42.50	0.81
Sept. 17	1	2115	50	78	27.12	0.64
Sept. 20	1	2184	40	70	31.20	0.57
Sept. 21	1	1807	26	69	26.19	0.38
Sept. 22	1	1183	17	43*	27.51	0.40
Sept. 23	1	141	2	43	3.28	0.05
Sept. 24	1	1437	17	41	35.05	0.41
Sept. 27	1	982	20	39	25.18	0.51
Sept. 28	1	1352	23	38	35.58	0.61
Sept. 29	1	0	0	37	0	0
Sept. 30	1	1712	21	37	46.27	0.57
Oct. 1	1	1179	17	35	33.69	0.49
Oct. 4	1	1499	27	33	45.42	0.82
Cladophora						
Aug. 25	1	49	3	157	0.31	0.02
Aug. 26	1	0	0	156	0	0
Aug. 27	1	0	0	154	0	0
Aug. 28	1	0	0	154	0	0
Aug. 31	1	21	1	150	0.14	0.01
Sept. 1	1	0	0	150	0	0
Sept. 8	1	5	1	119	0.04	0.01
Sept. 9	1	0	0	119	0	0
Sept. 10	1	0	0	117	0	0
Sept. 14	1	0	0	91*	0	0
Sept. 15	1	0	0	88	0	0
Sept. 16	1	0	0	87	0	0
Sept. 17	1	0	0	87	0	0
Sept. 20	1	0	0	82	0	0
Sept. 21	1	0	0	78	0	0
Sept. 22	1	0	0	52*	0	0
Sept. 23	1	0	0	50	0	0
Sept. 24	1	0	0	47	0	0

(continued)

TABLE 13, continued.

	No. Days	No. Eggs	No. egg Masses	No. Snails	Eggs/ snails/day	Egg masses/ snails/day
Sept. 27	1	0	0	41	0	0
Sept. 28	1	0	0	39	0	0
Sept. 29	1	0	0	36	0	0
Sept. 30	1	0	0	33	0	0
Oct. 1	1	0	0	32	0	0
Oct. 4	1	0	0	28	0	0

*Snails removed for respiration experiments

TABLE 14. NUMBERS, WEIGHTS, AND CALORIC CONTENT OF SNAIL EGGS, 1978 EXPERIMENT

Sample	Date	Diet	# Eggs	mg/egg	cal/mg AFDW
245	1/27/78	Periphyton	275	0.034	4.966
234	1/27/78	Periphyton	244	0.049	4.327 $\bar{X}$ 4.647
242	2/6/78	*Cladophora* + Epiphytes	427	0.010	4.718
232	1/27/78	*Cladophora*	192	0.023	4.215
233	1/27/78	*Cladophora*	170	0.033	3.998 $\bar{X}$ 4.106
231	1/27/78	Starved	175	0.04	4.075
235	1/30/78	Starved	408	0.022	4.087 $\bar{X}$ 4.081

TABLE 15. SNAIL FECES, 1977-1978 EXPERIMENTS

	Date	Dry Weight (mg)	Ash Free Dry Weight (mg)	% Ash Free Dry Weight
Periphyton				
1978	Jan. 25	25.34	21.54	85.0
	Jan. 26	47.28	38.94	82.4
	Jan. 29	51.70	43.12	83.4
	Jan. 30	35.80	32.20	89.9
	Feb. 3	21.90	18.16	82.9
	Feb. 4	36.52	30.44	83.4
	$\overline{X}$	34.423	30.733	84.5
	SD	11.708	6.650	2.786
	N	6	6	6
Cladophora + Epiphytes				
1978	Jan 25	12.18	8.68	71.3
	Jan. 27	21.92	15.06	68.7
	Jan. 30	28.34	21.66	76.4
	Jan. 31	25.98	17.94	69.1
	Feb. 8	27.62	22.92	83.0
	$\overline{X}$	23.208	17.252	73.70
	SD	6.648	5.708	6.035
	N	5	5	5
Cladophora				
1978	Jan. 25	7.48	3.84	51.3
	Jan. 30	15.62	10.72	68.6
	Jan. 30	7.06	4.36	61.8
	$\overline{X}$	10.053	6.307	60.567
	SD	4.825	3.831	8.716
	N	3	3	3

(continued)

TABLE 15, continued.

	Date	Dry Weight (mg)	Ash Free Dry Weight (mg)	% Ash Free Dry Weight
Starved				
1978	Jan. 25, 27	17.70	13.54	76.5
	Jan. 30, 31	6.62	5.00	75.5
	Feb. 1, 2	5.92	4.06	68.6
	$\overline{X}$	10.08	7.533	73.533
	SD	6.608	5.223	4.302
	N	3	3	3
Periphyton				
1977	May 23	42.20	37.66	89.2
	May 25	43.80	39.88	91.1
	June 2	41.48	36.98	89.2
	June 13	67.98	61.50	90.5
	July 8	26.60	23.22	87.3
	July 13	52.88	36.88	69.7
	$\overline{X}$	46.157	39.353	86.167
	SD	14.41	12.375	8.173
	N	6	6	6
Cladophora				
1977	May 25	3.08	1.84	59.7
	May 27	3.70	1.78	48.1
	June 2	9.42	5.22	55.4
	$\overline{X}$	5.4	2.947	54.4
	SD	3.495	1.969	5.864
	N	3	3	3

TABLE 16. UTILIZATION OF CALORIES INGESTED PER SNAIL, 1978 EXPERIMENT

	Periphyton	Cladophora + Epiphytes	Cladophora	Starved
January 25				
(Start of Experiment)				
Tissue $\overline{X}$ cal./snail	23.78	23.78	23.78	23.78
January 31				
(6-day period)				
Tissue $\overline{X}$ cal./snail	38.26	19.50	16.86	14.90
Gain or loss	+14.48	- 4.28	- 6.92	- 8.88
Respiration $\overline{X}$ cal./snail	14.83	5.33	4.18	3.17
Egg production (cal.)	23.04	3.26	1.39	1.382
Total calories assimilated	52.35	12.87	12.49	13.43
% wt. gain or loss	27.7	(33.3)	55.4	66.1
% respiration	28.3	41.4	33.5	23.6
% egg production	44.0	25.3	11.1	10.3
Calories ingested	257.06	53.68	19.4	-
% wt. gain or loss	5.6	9.9	35.7	-
% respiration	5.8	9.9	30.1	-
% egg production	6.2	5.2	14.5	-
% intake assimilated	17.6	-	-	-

TABLE 17. UTILIZATION OF CALORIES ASSIMILATED PER SNAIL, 1977 EXPERIMENTS

	Periphyton	Cladophora
May 19		
(Start of Experiment)		
Tissue $\overline{X}$ cal./snail	4.23	4.23
June 2		
Tissue $\overline{X}$ cal./snail	50.04	3.81
Gain or loss	45.81	(-0.42)
Respiration $\overline{X}$ cal./snail	39.48	1.34
Total calories used in growth and respiration	85.29	1.76
% in tissue growth or loss	53.7	(23.9)
% in respiration	46.3	76.1
June 2-16		
Tissue $\overline{X}$ cal./snail	72.1	3.079
Gain or loss	22.07	- 0.63
Respiration	47.38	1.18
Total calories used in growth and respiration	69.45	1.81
% in tissue gain or loss	31.8	(34.8)
% in respiration	68.2	65.2
June 16-30		
Tissue $\overline{X}$ cal./snail	121.91	1.95
Gain or loss	49.8	(- 1.13)
Respiration	98.6	0.34
Total calories used in growth and respiration	148.4	1.47
% in tissue gain or loss	33.6	(76.9)
% in respiration	66.4	23.1
June 30-July 14		
Tissue $\overline{X}$ cal./snail	143.45	2.054
Gain or loss	21.54	0.102
Respiration	97.47	3.248
Total calories used in growth and respiration	119.01	3.350
% in tissue gain or loss	18.1	3
% in respiration	81.9	97

TABLE 18. INDIVIDUAL WEIGHTS AND CALORIC VALUES, 1977 CRAYFISH EXPERIMENTS

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Start of Experiment July 18						
Aquarium 7 ♀	1.08	1.14	0.808	0.853	3635[a]	3837
	0.93	1.54	0.697	1.152	3136	5183
	1.65	0.89	1.234	0.665	5552	2992
	1.10	1.48	0.823	1.107	3703	4980
	1.39	1.62	1.040	1.212	4679	5453
	1.33	0.86	0.995	0.643	4476	2893
	1.22	1.27	0.913	0.950	4107	4274
	1.40	1.17	1.047	0.875	4710	3936
	1.39	1.02	1.040	0.763	4679	3433
	1.73	1.27	1.294	0.950	5821	4274
$\overline{X}$	1.27		0.953		4288	
SD	0.25		0.190		856	
N	20		20		20	
Aquarium 8 ♂	1.58	1.18	1.071	0.800	4174[b]	3118
	2.46	2.46	1.668	1.668	6500	6500
	2.74	1.86	1.858	1.261	7241	4914
	1.35	1.08	0.915	0.732	3566	2853
	2.06	2.38	1.397	1.614	5444	6290
	1.93	2.37	1.309	1.607	5101	6262
	2.25	0.87	1.526	0.590	5947	2299
	1.60	3.24	1.085	2.197	4228	8562
	1.84	1.51	1.248	1.024	4863	3991
	2.01	1.19	1.363	0.807	5312	3145
$\overline{X}$	1.90		1.287		5016	
SD	0.60		0.416		1621	
N	20		20		20	
Aquarium 9 ♀	1.01	1.14	0.666	0.751	2996	3379
	1.27	0.98	0.837	0.646	3765	2906
	1.36	0.89	0.896	0.587	4031	2641
	0.81	0.80	0.534	0.527	2402	2371
	1.23	1.20	0.811	0.791	3649	3559
	1.18	1.24	0.778	0.817	3500	3676
	1.48	1.11	0.975	0.731	3577	3289
	1.46	1.43	0.962	0.942	4328	4237
	0.88	0.81	0.580	0.534	2609	2402
	1.08	1.19	0.712	0.784	3203	3527
$\overline{X}$	1.13		0.743		3302	
SD	0.22		0.143		600	
N	20		20		20	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 10 ♂	1.63	1.38	1.074	0.909	4185	3542
	1.41	1.52	0.929	1.002	3620	3905
	1.28	1.64	0.844	1.081	3289	4213
	1.87	1.63	1.232	1.074	4801	4185
	1.41	1.64	0.929	1.081	3620	4213
	1.70	1.91	1.120	1.259	4365	4906
	1.67	1.47	1.101	0.969	4291	3776
	1.71	2.33	1.127	1.535	4392	5982
	1.32	1.69	0.870	1.114	3390	4341
	1.28	2.99	0.844	1.970	3289	7677
$\overline{X}$	1.67		1.103		4299	
SD	0.40		0.261		1019	
N	20		20		20	
Aquarium 11 ♂	1.58	1.41	1.013	0.904	3948	3523
	1.72	1.45	1.103	0.929	4298	3620
	1.54	1.86	0.987	1.192	3846	4645
	1.79	1.34	1.147	0.859	4470	3348
	1.85	1.41	1.186	0.904	4622	3523
	0.95	1.38	0.609	0.885	2373	3449
	2.06	1.22	1.320	0.782	5144	3047
	1.32	1.34	0.846	0.859	3297	3348
	1.36	1.43	0.872	0.917	3398	3574
	1.36	1.08	0.872	0.692	3398	2697
$\overline{X}$	1.47		0.944		3678	
SD	0.27		0.175		680	
N	20		20		20	
Aquarium 12 ♀	0.83	1.04	0.561	0.703	2524	3163
	1.40	1.30	0.946	0.879	4256	3954
	1.32	0.79	0.892	0.534	4013	2402
	0.72	0.92	0.487	0.622	2191	2798
	0.78	0.85	0.527	0.575	2371	2587
	1.42	0.92	0.960	0.622	4319	2798
	1.76	0.95	1.190	0.642	5354	2888
	1.97	0.59	1.332	0.399	5992	1795
	1.06	1.22	0.717	0.825	3226	3712
	0.98	1.14	0.662	0.771	2978	3469
$\overline{X}$	1.10		0.739		3340	
SD	0.35		0.240		1066	
N	20		20		20	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Periphyton						
July 28						
Aquarium 7 - ♀	0.86	1.15	0.643	0.860	2893[a]	3869
	1.04	0.80	0.778	0.598	3500	2690
	1.07	1.82	0.800	1.361	3599	6123
	1.36	1.71	1.017	1.279	4575	5754
	1.22	1.63	0.913	1.219	4107	5484
	1.36	0.98	1.017	0.733	4575	3298
	1.75	1.52	1.309	1.137	5889	5115
	1.25	1.44	0.935	1.077	4206	4845
	1.68	1.11	1.257	0.830	5655	3734
X̄	1.32		0.987		4440	
SD	0.32		0.236		1063	
N	18		18		18	
Aquarium 8 - ♂	1.61	1.26	1.092	0.854	4256[b]	3328
	2.56	1.95	1.736	1.322	6765	5152
	2.75	1.13	1.865	0.766	7268	2985
	1.35	2.41	0.915	1.634	3566	6368
	2.08	1.47	1.410	0.994	5495	3874
	2.01	2.44	1.363	1.650	5312	6430
	1.62	3.06	1.098	2.075	4279	8086
	1.67	2.42	1.132	1.640	4411	6391
	2.04		1.383		5390	
X̄	1.99		0.349		5256	
SD	0.56		0.377		1471	
N	17		17		17	
August 18						
Aquarium 7 - ♀	1.12	1.26	0.838	0.942	3770	4238
	0.95	1.67	0.711	1.249	3199	5619
	1.01	1.68	0.755	1.257	3397	5655
	1.52	1.47	1.137	1.100	5115	4949
	1.32	1.58	0.987	1.182	4440	5318
	1.69	1.87	1.264	1.399	5686	6294
	1.32	1.13	0.987	0.845	4440	3801
	1.81		1.354		6091	
X̄	1.43		1.067		4801	
SD	0.29		0.219		987	
N	15		15		15	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 8 - ♂	1.51	1.66	1.024	1.125	3991	4384
	2.41	1.77	1.634	1.200	6368	4676
	2.64	1.86	1.790	1.264	6976	4926
	1.32	2.36	0.895	1.600	3488	6235
	2.08	3.06	1.410	2.075	5495	8086
	2.28	1.43	1.546	0.970	6025	3780
	1.64	2.42	1.112	1.641	4333	6395
$\overline{X}$	2.03		1.377		5368	
SD	0.51		0.348		1356	
N	14		14		14	
August 29						
Aquarium 7 - ♀	1.06	1.22	0.793	0.913	3568	4107
	1.67	2.15	1.249	1.608	5620	7234
	0.99	1.49	0.741	1.115	3334	5016
	1.49	1.85	1.115	1.384	5016	6226
	1.67	1.89	1.249	1.414	5619	6361
	1.35		1.010		4544	
$\overline{X}$	1.53		1.14		5149	
SD	0.36		0.270		1212	
N	11		11		11	
Aquarium 8 - ♂	2.55	1.66	1.730	1.125	6742	4384
	3.03	1.77	2.054	1.200	8004	4676
	1.35	1.88	0.914	1.275	3562	4969
	2.16	2.43	1.464	1.648	5705	6422
	2.37	3.31	1.607	2.244	6262	8745
$\overline{X}$	2.25		1.53		5947	
SD	0.62		0.417		1625	
N	10		10		10	
September 8						
Aquarium 7 - ♀	0.99	1.71	0.741	1.279	3334	5754
	1.63	1.16	1.219	0.868	5484	3905
	0.98	1.44	0.733	1.077	3298	4845
	1.48	1.58	1.107	1.182	4980	5318
	1.64	1.88	1.227	1.406	5520	6325
	1.34	1.11	1.002	0.830	4508	3734
$\overline{X}$	1.41		1.055		4751	
SD	0.30		0.221		997	
N	12		12		12	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 8 - ♂	2.48	1.64	1.681	1.112	6551	4333
	2.68	1.75	1.817	1.187	7081	4626
	1.14	1.75	0.773	1.187	3012	4626
	2.10	3.02	1.424	2.048	5549	7981
	2.24	1.43	1.519	0.970	5920	3780
$\overline{X}$	2.02		1.37		5346	
SD	0.59		0.399		1554	
N	10		10		10	
September 19						
Aquarium 7 - ♀	1.03	1.72	0.770	1.287	3464	5790
	1.63	1.20	1.219	0.898	5484	4040
	0.98	1.43	0.733	1.070	3298	4814
	1.45	1.52	1.085	1.137	4881	5115
	1.61	1.85	1.204	1.384	5417	6226
	1.64	1.12	1.227	0.838	5520	3770
$\overline{X}$	1.43		1.07		4818	
SD	0.29		0.214		961	
N	12		12		12	
Aquarium 8- ♂	2.46	1.68	1.668	1.139	6500	4439
	2.55	2.32	1.729	1.573	6738	6130
	2.06	2.97	1.397	2.014	5444	7849
	2.23	1.41	1.512	0.956	5892	3726
	1.60		1.085		4228	
$\overline{X}$	2.14		1.34		5661	
SD	0.51		0.297		1335	
N	9		9		9	
September 29						
Aquarium 7 - ♀	1.04	1.74	0.778	1.302	3500	5857
	1.61	1.21	1.204	0.905	5417	4071
	1.01	1.58	0.755	1.182	3397	5318
	1.48	1.88	1.107	1.406	4980	6325
	1.65	1.14	1.234	0.853	5552	3837
	1.30		0.972		4373	
$\overline{X}$	1.42		1.063		4834	
SD	0.30		0.222		710	
N	11		11		11	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 8 - ♂	2.45	1.70	1.661	1.153	6473	4493
	2.54	2.32	1.722	1.573	6711	6130
	2.24	2.93	1.519	1.987	5920	7743
	1.58	1.41	1.071	0.956	4174	3726
$\overline{X}$	2.15		1.455		5671	
SD	0.53		0.359		1399	
N	8		8		8	
October 13						
Aquarium 7 - ♀	1.00	1.26	0.748	0.942	3365	4238
	1.58	1.67	1.182	1.249	5318	5619
	0.96	1.81	0.718	1.353	3230	6087
	1.44	1.12	1.077	0.838	4845	3770
	1.57		1.174		5282	
$\overline{X}$	1.38		1.031		4639	
SD	0.30		0.229		1031	
N	9		9		9	
Aquarium 8 - ♂	2.42	1.54	1.641	1.044	6395	4068
	2.50	2.90	1.695	1.967	6605	7665
	2.18	1.37	1.478	0.929	5760	3620
$\overline{X}$	2.15		1.459		5686	
SD	0.59		0.400		1559	
N	6		6		6	
November 15						
Aquarium 7 - ♀	1.00	1.56	0.748	1.167	3365	5250
	1.55	1.24	1.159	0.928	5214	4175
	0.88	1.79	0.658	1.339	2960	6024
	1.41	1.07	1.055	0.800	4746	3599
$\overline{X}$	1.31		0.982		4416	
SD	0.32		0.237		1068	
N	8		8		8	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 8 - ♂	2.37	1.50	1.609	1.017	6270	3963
	2.48	2.86	1.681	1.939	6551	7556
	2.15	1.33	1.458	0.902	5682	3515
$\overline{X}$	2.12		1.434		5590	
SD	0.59		0.401		1563	
N	6		6		6	
December 7						
Aquarium 7 - ♀	0.99	1.56	0.741	1.167	3334	5250
	1.56	1.44	1.167	1.077	5250	4845
	0.88	1.79	0.658	1.339	2960	6024
	1.41		1.055		4746	
$\overline{X}$	1.38		1.029		4630	
SD	0.33		0.244		1099	
N	7		7		7	
Aquarium 8 - ♂	2.45	1.51	1.661	1.024	6473	3991
	2.15	1.34	1.458	0.909	5682	3542
$\overline{X}$	1.86		1.263		4922	
SD	0.52		0.355		1385	
N	4		4		4	
December 14						
Aquarium 7 - ♀	0.99	1.55	0.741	1.159	3334	5214
	1.56	1.33	1.167	0.995	5250	4476
	0.88	1.78	0.658	1.331	2960	5988
	1.40		1.047		4710	
$\overline{X}$	1.36		1.014		4562	
SD	0.32		0.241		1083	
N	7		7		7	
Aquarium 8 - ♂	2.48	1.33	1.681	0.902	6551	3515
	2.15		1.458		5682	
$\overline{X}$	1.99		1.347		5249	
SD	0.59		0.401		1564	
N	3		3		3	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Cladophora + Epiphytes						
July 28						
Aquarium 9 ♀	1.39	1.29	0.916	0.850	3593[c]	3334
	1.57	1.42	1.035	0.936	4060	3671
	1.20	0.83	0.791	0.547	3103	2146
	1.45	1.13	0.956	0.745	3750	2922
	1.39	1.31	0.916	0.863	3593	3385
	0.99	0.81	0.652	0.534	2557	2095
	1.12	1.36	0.738	0.896	2895	3515
	0.92	1.04	0.606	0.685	2377	2687
	0.90	1.14	0.593	0.751	2326	2946
	0.83	1.21	0.547	0.797	2146	3126
$\overline{X}$	1.17		0.768		3011	
SD	0.23		0.153		599	
N	20		20		20	
Aquarium 10 ♂	1.62	1.54	1.068	1.015	3955[d]	3759
	1.37	1.64	0.903	1.081	3344	4003
	1.27	1.64	0.837	1.081	3100	4003
	1.90	1.42	1.252	0.936	4636	3466
	1.44	1.93	0.949	1.272	3514	4710
	1.75	2.38	1.153	1.568	4270	5807
	1.70	2.97	1.120	1.957	4148	7247
	1.32	1.45	0.870	0.956	3222	3540
	1.26	1.68	0.830	1.107	3074	4099
	1.36	1.11	0.896	0.731	3318	2707
$\overline{X}$	1.64		1.079		3996	
SD	0.43		0.280		1038	
N	20		20		20	
August 18						
Aquarium 9 ♀	1.13	0.78	0.745	0.514	2922	2016
	1.32	1.34	0.870	0.883	3413	3464
	1.47	1.60	0.969	1.054	3801	4134
	0.96	1.14	0.663	0.751	2601	2946
	1.19	1.26	0.784	0.830	3075	3256
	1.45	1.45	0.956	0.956	3750	3750
	1.45	0.89	0.956	0.587	3750	2303
	1.38	0.93	0.909	0.613	3566	2404
	1.27		0.837		3283	
$\overline{X}$	1.24		0.815		3202	
SD	0.24		0.155		600	
N	17		17		17	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 10 ♂	1.60	1.55	1.054	1.021	3903	3781
	1.30	1.59	0.857	1.048	3174	3881
	1.86	1.40	1.226	0.923	4540	3418
	1.45	1.86	0.956	1.226	3540	4540
	1.67	1.48	1.101	0.975	4077	3611
	1.70	2.27	1.120	1.496	4148	5540
	1.26	1.64	0.830	1.081	3074	4003
	1.32	2.90	0.870	1.911	3222	7077
	1.49		0.982		3637	
X̄	1.67		1.127		4068	
SD	0.40		0.285		982	
N	17		17		17	
August 29						
Aquarium 9 ♀	1.13	0.82	0.745	0.540	2922	2118
	1.31	1.06	0.863	0.698	3385	2738
	1.50	1.35	0.989	0.890	3879	3491
	1.03	1.16	0.679	0.764	2663	2997
	1.31	1.27	0.863	0.837	3385	3283
	1.50	1.45	0.989	0.956	3879	3750
	1.41	0.92	0.929	0.606	3644	2377
	0.96	1.23	0.633	0.811	2483	3181
	1.26		0.830		3256	
X̄	1.22		0.801		3143	
SD	0.21		0.135		531	
N	17		17		17	
Aquarium 10 ♂	1.55	1.48	1.021	0.975	3781	3611
	1.29	1.63	0.850	1.074	3148	3977
	2.20	1.44	1.500	0.949	5555	3514
	1.70	1.92	1.120	1.265	4148	4685
	1.73	1.44	1.140	0.949	4222	3514
	1.12	1.65	0.738	1.087	2733	4025
	1.58	2.63	1.041	1.733	3855	6418
X̄	1.67		1.103		4084	
SD	0.38		0.256		950	
N	14		14		14	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Sept. 8						
Aquarium 9 ♀	1.23	0.90	0.811	0.593	3181	2326
	1.50	1.17	0.989	0.771	3879	3024
	1.69	1.57	1.114	1.035	4370	4060
	1.15	1.29	0.758	0.850	2973	3334
	1.40	1.44	0.923	0.949	3620	3722
	1.69	1.53	1.114	1.008	4370	3954
	1.51	1.01	0.995	0.666	3903	2612
	1.40	1.08	0.923	0.712	3620	2793
$\overline{X}$	1.35		0.888		3483	
SD	0.24		0.157		617	
N	16		16		16	
Aquarium 10 ♂	1.55	1.73	1.021	1.140	3781	4222
	1.25	1.71	0.824	1.127	3051	4173
	1.83	1.67	1.206	1.101	4466	4077
	1.70	1.91	1.120	1.259	4148	4662
	1.36	1.39	0.896	0.916	3318	3392
	1.17	1.64	0.771	1.081	2855	4003
	1.53	3.07	1.008	2.023	3733	7492
	1.52		1.002		3711	
$\overline{X}$	1.67		1.100		4072	
SD	0.44		0.290		1073	
N	15		15		15	
Sept. 19						
Aquarium 9 ♀	1.10	1.21	0.725	0.797	2844	3126
	1.26	0.77	0.830	0.507	3256	1989
	1.42	1.00	0.936	0.659	3671	2585
	0.97	1.00	0.639	0.659	2506	2585
	1.24	1.24	0.817	0.817	3205	3205
	1.43	1.37	0.942	0.903	3695	3542
	1.33	0.87	0.876	0.573	3436	2248
	1.33	0.90	0.876	0.593	3436	2326
$\overline{X}$	1.15		0.670		2978	
SD	0.21		0.138		543	
N	16		16		16	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 10 ♂	1.55	1.53	1.021	1.008	3781	3733
	1.25	1.55	0.824	1.021	3051	3781
	1.84	1.32	1.213	0.870	4492	3222
	1.64	1.85	1.081	1.219	4003	4514
	1.66	1.39	1.094	0.916	4051	3392
	1.22	1.60	0.804	1.054	2977	3903
	1.34	2.83	0.883	1.865	3270	6906
	1.46		0.962		3562	
$\overline{X}$	1.60		1.060		3909	
SD	0.39		0.257		951	
N	15		15		15	
Sept. 29						
Aquarium 9 ♀	1.10	1.22	0.725	0.804	2844	3154
	1.24	0.76	0.817	0.501	3205	1965
	1.42	1.00	0.936	0.659	3671	2585
	0.99	1.22	0.652	0.804	2557	3154
	1.25	1.37	0.824	0.903	3232	3542
	1.41	0.87	0.929	0.573	3644	2248
	1.36	0.89	0.896	0.587	3515	2303
	1.36		0.896		3515	
$\overline{X}$	1.16		0.767		3005	
SD	0.22		0.142		553	
N	15		15		15	
Aquarium 10 ♂	1.55	1.52	1.021	1.002	3781	3711
	1.24	1.55	0.817	1.021	3025	3781
	1.82	1.34	1.199	0.883	4440	3270
	1.66	1.86	1.094	1.226	4051	4540
	1.67	1.40	1.101	0.923	4077	3418
	1.22	1.62	0.804	1.068	2977	3955
	1.31	2.81	0.863	1.852	3196	6858
	1.43		0.942		3488	
$\overline{X}$	1.60		1.054		3904	
SD	0.39		0.255		944	
N	15		15		15	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Oct. 13						
Aquarium 9 ♀	1.10	1.33	0.752	0.883	2950	3464
	1.23	1.34	0.811	0.784	3181	3075
	1.42	1.19	0.936	0.639	3671	2506
	0.97	0.97	0.639	0.883	2506	3464
	1.22	1.34	0.804	0.573	3154	2248
	1.43	0.87	0.942	0.567	3695	2224
	0.86		0.876		3436	
$\overline{X}$	1.18		0.774		3044	
SD	0.20		0.133		520	
N	13		13		13	
Aquarium 10 ♂	1.51	1.43	0.995	0.942	3685	3488
	1.21	1.49	0.797	0.982	2951	3636
	1.80	1.53	1.186	1.008	4392	3733
	1.62	1.37	1.068	0.903	3955	3344
	1.21	1.57	0.797	1.035	2951	3833
	1.31	2.75	0.863	1.812	3196	6710
$\overline{X}$	1.57		1.032		3823	
SD	0.41		0.270		1000	
N	12		12		12	
Oct. 28						
Aquarium 9 ♀	1.07	1.30	0.705	0.837	2765	3283
	1.22	1.27	0.804	0.778	3154	3052
	1.41	1.18	0.929	0.633	3644	2483
	0.95	0.96	0.626	0.863	2455	3385
	1.20	1.31	0.791	0.554	3103	2173
	1.39	0.84	0.916	0.573	3593	2248
	0.87		0.857		3362	
$\overline{X}$	1.15		0.759		2977	
SD	0.19		0.128		502	
N	13		13		13	
Aquarium 10 ♂	1.48	1.27	0.975	0.837	3610	3100
	1.19	1.46	0.784	0.962	2903	3562
	1.77	1.33	1.167	0.876	4321	3244
	1.60	1.55	1.054	1.021	3903	3781
	1.18	2.73	0.778	1.799	2881	6662
$\overline{X}$	1.56		0.925		3797	
SD	0.45		0.132		1106	
N	10		10		10	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Dec. 7						
Aquarium 9 ♀	0.94	1.17	0.619	0.771	2428	3024
	1.19	1.32	0.784	0.870	3075	3413
	1.36	0.84	0.896	0.554	3515	2173
	1.27	0.86	0.837	0.567	3283	2224
$\bar{X}$	1.12		0.737		2892	
SD	0.21		0.138		540	
N	8		8		8	
Aquarium 10 ♂	1.15		0.758		2807	
	1.24		0.817		3025	
$\bar{X}$	1.20		0.788		2916	
SD	0.06		0.042		154	
N	2		2		2	
Dec. 14						
Aquarium 9 ♀	0.94	1.17	0.619	0.771	2428	3024
	1.20	0.84	0.791	0.554	3103	2173
	1.36	0.86	0.896	0.567	3515	2224
	1.27		0.837		3283	
$\bar{X}$	1.09		0.719		2821	
SD	0.21		0.138		539	
N	7		7		7	
Aquarium 10 ♂	1.16		0.764		2829	
	1.26		0.830		3074	
$\bar{X}$	1.21		0.797		2951	
SD	0.07		0.047		173	
N	2		2		2	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Cladophora						
July 28						
Aquarium 11-♂	1.90	1.50	1.218	0.962	3959[f]	3127
	1.73	1.49	1.109	0.955	3604	3104
	1.45	1.78	0.929	1.141	3019	3708
	1.71	1.36	1.096	0.872	3562	2834
	1.91	1.47	1.224	0.942	3978	3062
	0.92	1.15	0.590	0.737	1918	2395
	2.14	1.31	1.372	0.840	4459	2730
	1.17	1.38	0.750	0.885	2438	2876
	1.34	1.44	0.859	0.923	2792	3000
	1.46	1.07	0.936	0.699	3042	2272
$\overline{X}$	1.49		0.952		3074	
SD	0.30		0.194		609	
N	20		20		29	
Aquarium 12-♀	1.16	0.82	0.784	0.554	2949[e]	2084
	0.70	0.84	0.473	0.568	1779	2136
	1.12	1.45	0.757	0.980	2847	3686
	1.67	1.14	1.129	0.771	4246	2900
	1.94	0.74	1.311	0.500	4931	1881
	1.01	0.84	0.683	0.568	2569	2136
	0.95	1.39	0.642	0.940	2415	3535
	0.81	0.80	0.548	0.541	2061	2035
	1.29	1.07	0.872	0.723	3280	2719
	1.07	0.59	0.723	0.399	2719	1501
$\overline{X}$	1.07		0.723		2720	
SD	0.34		0.232		871	
N	20		20		20	
August 18						
Aquarium 11-♂	1.58	1.43	1.013	0.917	3292	2980
	1.68	1.45	1.077	0.929	3500	3019
	1.47	1.80	0.942	1.154	3062	3751
	1.82	1.35	1.167	0.865	3793	2811
	1.87	1.41	1.199	0.904	3897	2938
	0.91	1.22	0.583	0.782	1895	2542
	2.09	1.35	1.340	0.865	4355	2811
	1.31	1.41	0.840	0.904	2730	2938
	1.38	1.04	0.885	0.667	2876	2168
	1.38		0.855		2779	
	1.47		0.943		3060	
	0.29		0.183		598	
	19		19		19	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
August 18						
Aquarium 12- ♀	1.06	0.86	0.717	0.581	2697	2185
	1.21	0.86	0.818	0.581	3076	2185
	1.28	0.76	0.865	0.514	3253	1933
	0.78	0.86	0.527	0.581	1982	2185
	1.38	0.89	0.933	0.602	3509	2264
	1.45	0.56	0.980	0.379	3686	1425
	1.67	1.15	1.129	0.777	4246	2922
	1.99	1.15	1.345	0.777	5059	2922
	1.07		0.723		2719	
	1.12		0.755		2838	
	0.35		0.244		919	
	17		17		20	
August 29						
Aquarium 11 - ♂	1.57	1.80	1.006	1.154	3270	3751
	1.46	1.37	0.936	0.878	3042	2854
	1.81	1.40	1.160	0.897	3770	2915
	1.83	1.22	1.173	0.782	3812	2542
	2.34	1.35	1.500	0.865	4875	2811
	1.39	1.44	0.891	0.923	2896	3000
	1.46	1.06	0.936	0.679	3042	2207
	1.48		0.949		3084	
	1.53		0.982		3191	
	0.31		0.197		643	
	15		15		15	
Aquarium 12 - ♀	1.14	0.90	0.771	0.608	2900	2287
	1.24	0.89	0.838	0.602	3152	2264
	1.32	0.77	0.892	0.521	3355	1959
	0.83	0.89	0.561	0.602	2110	2264
	1.35	0.92	0.913	0.622	3434	2339
	1.43	0.55	0.967	0.372	3637	1399
	1.72	1.20	1.163	0.811	4374	3050
	2.08	1.15	1.406	0.777	5288	2922
	1.12		0.751		2825	
	1.15		0.775		2915	
	0.37		0.251		944	
	17		17		17	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Sept. 8						
Aquarium 11 ♂	1.58	1.80	1.013	1.154	3292	3751
	1.69	1.02	1.083	0.654	3520	2126
	1.76	1.37	1.128	0.878	3666	2854
	1.85	1.21	1.186	0.776	3855	2522
	2.02	1.34	1.295	0.859	4209	2792
	1.36	1.44	0.872	0.923	2834	3000
	1.41	1.04	0.904	0.667	2938	2168
	1.43		0.917		2980	
$\overline{X}$	1.49		1.000		3100	
SD	0.29		0.293		609	
N	15		15		15	
Aquarium 12 ♀	1.26	1.33	0.852	0.899	3204	3381
	1.47	0.89	0.994	0.602	3738	2264
	1.11	0.45	0.750	0.304	2821	1143
	0.89	1.04	0.602	0.703	2264	2644
	1.60	0.90	1.082	0.608	4069	2287
	1.38	0.78	0.933	0.527	3509	1982
	1.69	1.34	1.142	0.906	4295	3407
	1.76	1.33	1.190	0.899	4476	3381
	1.28		0.865		3253	
$\overline{X}$	1.21		0.815		3066	
SD	0.35		0.235		885	
N	17		17		17	
Sept. 19						
Aquarium 11 ♂	1.55	1.78	0.994	1.141	3231	3708
	1.67	1.33	1.070	0.853	3478	2772
	1.79	1.39	1.147	0.891	3728	2896
	1.81	1.22	1.160	0.782	3770	2542
	2.02	1.34	1.295	0.859	4209	2792
	1.34	1.40	0.859	0.897	2792	2915
	1.40	1.03	0.897	0.660	2915	2145
	1.41		0.904		2938	
$\overline{X}$	1.50		0.960		3122	
SD	0.27		0.170		551	
N	15		15		15	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 12 ♀	1.10	1.28	0.744	0.865	2798	3253
	1.22	0.89	0.825	0.602	3103	2264
	1.09	0.90	0.737	0.608	2772	2287
	0.77	0.90	0.521	0.608	1959	2287
	1.40	0.76	0.946	0.514	3558	1933
	1.40	1.20	0.946	0.811	3558	3050
	1.67	1.09	1.129	0.737	4246	2772
	1.99		1.345		5059	
$\overline{X}$	1.18		0.800		2993	
SD	0.34		0.230		363	
N	15		15		15	
Sept. 29						
Aquarium 11 ♂	1.53	1.77	0.981	1.135	3188	3689
	1.66	1.30	1.064	0.833	3458	2707
	1.78	1.40	1.141	0.897	3708	2915
	1.82	1.34	1.167	0.859	3793	2792
	2.01	1.37	1.288	0.878	4186	2854
	1.35	1.04	0.865	0.667	2811	2168
	1.41		0.904		2938	
$\overline{X}$	1.52		0.975		3170	
SD	0.27		0.173		561	
N	13		13		13	
Aquarium 12 ♀	1.05	1.93	0.710	1.305	2670	4908
	1.18	0.87	0.798	0.588	3001	2211
	1.06	0.89	0.717	0.602	2697	2264
	0.77	0.87	0.521	0.588	1959	2211
	1.38	0.77	0.933	0.521	3509	1959
	1.38	1.13	0.933	0.764	3509	2873
	1.64	1.14	1.109	0.771	4171	2900
$\overline{X}$	1.15		0.776		2917	
SD	0.34		0.228		860	
N	14		14		14	
Oct 13						
Aquarium 11 ♂	1.52	1.38	0.974	0.885	3166	2876
	1.66	1.73	1.064	1.109	3458	3604
	1.75	1.30	1.122	0.833	3647	2707
	1.79	1.30	1.147	0.833	3728	2707
	1.98	1.36	1.269	0.872	4124	2834
	1.35	1.01	0.865	0.647	2811	2103
$\overline{X}$	1.51		0.968		3147	
SD	0.27		0.176		573	
N	12		12		12	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 12 ♀	1.06	1.93	0.717	1.305	2697	4908
	1.18	0.89	0.798	0.602	3001	2264
	1.07	0.87	0.723	0.588	2719	2211
	0.77	0.76	0.521	0.514	1959	1933
	1.37	1.15	0.926	0.777	3483	2922
	1.37	1.13	0.926	0.764	3483	2873
	1.64		1.109		4171	
$\overline{X}$	1.17		0.790		2971	
SD	0.34		0.230		868	
N	13		13		13	
Oct. 28						
Aquarium 11 ♂	1.48	1.34	0.949	0.859	3084	2792
	1.71	1.70	1.096	1.090	3562	3543
	1.93	1.27	1.237	0.814	4020	2646
	1.30	1.34	0.833	0.859	2707	2792
$\overline{X}$	1.51		0.967		3143	
SD	0.24		0.156		506	
N	8		8		8	
Aquarium 12 ♀	1.05	1.88	0.710	1.271	2670	4780
	1.15	0.86	0.777	0.581	2922	2185
	1.03	0.75	0.696	0.507	2618	1907
	0.68	1.13	0.460	0.764	1730	2873
	1.33	1.09	0.899	0.737	3381	2772
	1.33		0.899		3381	
$\overline{X}$	1.12		0.755		2838	
SD	0.33		0.222		835	
N	11		11		11	
Dec. 7						
Aquarium 11 ♂	1.46		0.936		3042	
	1.30		0.833		2707	
	1.33		0.853		2772	
$\overline{X}$	1.36		0.874		2840	
SD	0.09		0.055		178	
N	3		3		3	

(continued)

TABLE 18, continued.

	Dry Weight g		Ash Free Dry Weight g		Ash Free Dry Weight cal/crayfish	
Aquarium 12 ♀	1.03	0.96	0.696	0.649	2618	2441
	1.32	0.74	0.892	0.500	3355	1881
	1.89	1.10	1.278	0.744	4807	2798
$\bar{X}$	1.17		0.793		2983	
SD	0.40		0.270		1014	
N	6		6		6	
Dec. 14						
Aquarium 11 ♂	1.46		0.936		3042	
	1.28		0.820		2665	
$\bar{X}$	1.37		0.878		2854	
SD	0.13		0.082		267	
N	2		2		2	
Aquarium 12 ♀	1.02		0.690		2595	
	1.89		1.278		4807	
	0.83		0.561		2110	
	0.74		0.500		1881	
	1.10		0.744		2798	
$\bar{X}$	1.11		0.755		2838	
SD	0.45		0.308		1160	
N	5		5		5	

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[a] Periphyton (♀) - 4498.8 cal/g ($\bar{X}$)
[b] Periphyton (♂) - 3897 cal/g ($\bar{X}$)
[c] *Cladophora* + Epiphytes (♀) - 3922.49 cal/g ($\bar{X}$)
[d] *Cladophora* + Epiphtyes (♂) - 3703.18 cal/g ($\bar{X}$)
[e] *Cladophora* (♀) - 3761 ($\bar{X}$)
[f] *Cladophora* (♂) - 3250 ($\bar{X}$)

TABLE 19. DATA FOR BASIS OF CONVERSIONS FROM DRY WEIGHT TO CALORIES, 1977 CRAYFISH TISSUE EXPERIMENTS

	Dry Weight g	Ash Free Dry Weight g	% Ash Free Dry Weight	Ash Free Dry Weight cal/g	
Periphyton					
Aquarium No. 7 ♀					
Aug. 15	0.869	0.667	76.8	4104	
	0.955	0.660	69.1	4614	
Aug. 22	0.932	0.705	75.6	4630	
	0.934	0.721	77.2	4720	
Sept. 27	0.925	0.707	76.4	4571	
Oct. 10	0.924	0.678	73.4	4354	
$\overline{X}$			74.8	4498.8	
SD			3.1	228.7	
N			6	6	
Aquarium No. 8 ♂					
Aug. 15	0.955	0.669	70.1	4053	
	0.728	0.517	71.0	4003	
Aug. 22	0.779	0.525	67.4	3961	
	0.967	0.674	69.7	4041	
Sept. 1	0.568	0.361	63.6	3506	
Sept. 30	0.954	0.632	66.2		
	1.017	0.680	66.9	3820	
$\overline{X}$			67.8	3897	
SD			2.6	209	
N			7	6	
				Aquarium 9 - ♀ March 1	Aquarium 10 - ♂ March 3
Cladophora + Epiphytes					
Aquaria Nos. 9 & 10 ♀ & ♂					
Feb. 9, 1978	1.56	1.09	69.9	3472	3550
	2.31	1.40	60.6	4040	3855
	1.17	0.79	67.5	3941	
				3986	
				4170	
$\overline{X}$			66.0	3922	3703
SD			4.8	266	216
N			3	5	2

(continued)

TABLE 19. continued.

	Dry Weight g	Ash Free Dry Weight g	% Ash Free Dry Weight	Ash Free Dry Weight cal/g
Cladophora				
Aquarium No. 11 ♂				
Aug. 22	0.918	0.567	61.8	3208
	0.831	0.495	59.6	3358
Sept. 1	0.947	0.626	66.1	3413
Sept. 30	1.230	0.892	72.5	
Oct. 10	0.912	0.551	60.4	3022
$\overline{X}$			64.1	3250
SD			5.3	175.1
N			5	4
Aquarium No. 12 ♀				
Aug. 15	0.648	0.452	69.7	3844
	0.580	0.400	69.0	3906
Sept. 1	0.947	0.659	69.6	4031
Sept. 30	0.973	0.642	66.0	3664
Oct. 10	0.458	0.290	63.3	3361
$\overline{X}$			67.5	3761
SD			2.8	259.9
N			5	5

TABLE 20. RESPIRATION, 1977 CRAYFISH EXPERIMENTS

	Dry Weight (gm) Crayfish Tissue			ml O_2/ g crayfish/ hr	cal/ g crayfish/ hr *	cal/ crayfish/ hr	cal/ crayfish/ day
	$\bar{X}$	SD	N				
Periphyton							
Oct. 3-4							
Aquarium 7 ♀	1.49	0.33	10	0.488	2.29	3.41	81.84
Aquarium 8 ♂	2.02	0.54	7	0.349	1.64	3.31	79.44
Dec. 13-14							
Aquarium 7 ♀	1.36	0.32	7	0.152	0.71	0.97	23.17
Aquarium 8 ♂	1.99	0.59	3	0.057	0.27	0.54	12.96
Cladophora + Epiphytes							
Oct. 3-4							
Aquarium 9 ♀	1.30	0.10	10	0.246	1.16	1.51	36.24
Aquarium 10 ♂	1.44	0.40	10	0.227	1.07	1.54	36.96
Dec. 13-14							
Aquarium 9 ♀	1.09	0.21	7	0.080	0.38	0.41	9.84
Aquarium 10 ♂	1.19	0.07	2	0.059	0.28	0.33	7.92
Cladophora							
Oct. 3-4							
Aquarium 11 ♂	1.51	0.25	10	0.318	1.49	2.25	54.00
Aquarium 12 ♀	1.21	0.28	10	0.257	1.21	1.46	35.04
Dec. 13-14							
Aquarium 11 ♂	1.41	0.13	2	0.077	0.36	0.51	12.24
Aquarium 12 ♀	1.15	0.42	6	0.138	0.65	0.75	18.00

* 1 ml O_2 - 4.7 calories

TABLE 21. FOOD CONSUMPTION, 1977 CRAYFISH EXPERIMENTS

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray- fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day *
Periphyton							
July 19-27							
Aquarium 7 - ♀							
$\bar{X}$	138.33	58.65	2.93	13.901	20	0.695	2990
SD	41.21	17.47	0.87	4.141		0.207	890.7
N	9	9	9	9		9	9
Aquarium 8 - ♂							
$\bar{X}$	100.89	42.78	2.28	10.138	20 (7/19-22) 18 (7/23-27)	0.540	2323
SD	32.09	13.61	0.77	3.225		0.182	782.6
N	9	9	9	9		9	9
July 28-Aug. 17							
Aquarium 7 - ♀							
$\bar{X}$	161.82	68.61	3.74	16.261	19 (7/28-8/7) 18 (8/8-12) 17 (8/15-17)	0.833	3583.6
SD	43.55	18.46	1.04	4.376		0.312	1061.0
N	19	19	19	19		19	19
Aquarium 8 - ♂							
$\bar{X}$	83.84	35,55	2.30	8,425	16 (7/18-8/11) 15 (8/12) 13 (8/15-17)	0.549	2361.8
SD	27.49	11.65	0.79	2.762		0.199	856.0
N	19	19	19	19		19	19

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray-fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Aug. 18-28							
Aquarium 7 - ♀							
$\bar{X}$	82.14	34.83	2.51	8.254	15	0.595	2559.7
SD	34.51	14.63	1.00	3.468	(8/18)	0.237	1019.6
N	9	9	9	9	14	9	9
					(8/19-24)		
					13		
					(8/25-28)		
Aquarium 8 - ♂							
$\bar{X}$	43.67	18.51	1.42	4.388	13	0.340	1462.7
SD	22.41	9.50	0.73	2.252		0.174	745.3
N	9	9	9	9		9	9
Aug. 29-Sept. 8							
Aquarium 7 - ♀							
$\bar{X}$	43.38	18.39	1.47	4.359	13	0.348	1497
SD	9.37	3.97	0.31	0.942	(8/29-9/1)	0.073	314.7
N	8	8	8	8	12	8	8
					(9/4-8)		
Aquarium 8 - ♂							
$\bar{X}$	26.50	11.24	1.04	2.66	13	0.247	1062.5
SD	11.37	4.82	0.44	1.14	(8/29)	0.104	447.1
N	8	8	8	8	11	8	8
					(8/30-9/1)		
					10		
					(9/4-8)		
Sept. 9-19							
Aquarium 7- ♀							
$\bar{X}$	67.43	28.59	2.38	6.777	12	0.565	2430.6
SD	34.83	14.77	1.23	3.500		0.292	1255.3
N	10	10	10	10		10	10

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray-fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Aquarium 8 - ♂							
$\bar{X}$	29.38	12.46	1.25	2.952	10	0.297	1277.7
SD	18.84	7.989	0.79	1.894	(9/9-18)	0.188	809.2
N	10	10	10	10	9	10	10
					(9/19)		
Sept. 20-29 Aquarium 7 - ♀							
$\bar{X}$	51.68	21.91	1.83	5.193	12	0.433	1862.7
SD	20.19	8.56	0.71	2.029		0.169	727.4
N	8	8	8	8		8	8
Aquarium 8 - ♂							
$\bar{X}$	36.54	12.47	1.76	2.954	9	0.339	1458.4
SD	11.59	5.72	0.50	1.356	(9/20-26)	0.148	637.1
N	7	7	7	7	8	7	7
					(9/28-29)		
Sept. 30-Oct. 4 Aquarium 7 - ♀							
$\bar{X}$	57.73	24.48	2.04	5.802	12	0.484	2082.1
SD	6.62	1.73	0.24	0.666		0.055	238.4
N	3	3	3	3		3	3
Aquarium 8 - ♂							
$\bar{X}$	33.17	14.06	1.81	2.636	8	0.428	1841.2
SD	17.64	7.48	0.87	0.162	(9/30-10/1)	0.208	895.6
N					7		
					(10/4)		
Oct. 5-6 Aquarium 7 - ♀							
$\bar{X}$	39.20	16.62	1.39	3.939	12	0.315	1355.1
SD	22.77	9.66	0.80	2.290		0.172	821.7
N	2	2	2	2		2	2

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray- fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Aquarium 8 - ♂							
$\bar{X}$	11.85	5.03	0.72	1.191	7	0.170	731.3
SD	10.54	4.46	0.64	1.058		0.151	651.2
N	2	2	2	2		2	2
Cladophora + Epiphytes							
July 20-27							
Aquarium 9 - ♀							
$\bar{X}$	4.79	0.503	0.025	0.347	20	0.017	74.94
SD	0.5378	0.0565	0.0028	0.0388		0.0021	9.1063
N	8	8	8	8		8	8
Aquarium 10 - ♂							
$\bar{X}$	4.77	0.501	0.026	0.345	20	0.018	79.34
SD	0.8583	0.0902	0.0045	0.0621	(7/20-21)	0.0029	12.7849
N	8	8	8	8	19 (7/22-27)	8	8
July 28-Aug. 18							
Aquarium 9 - ♀							
$\bar{X}$	5.17	0.543	0.027	0.374	20	0.019	83.75
SD	1.2202	0.1284	0.0063	0.0883	(7/28-8/14)	0.0041	18.1150
N	19	19	19	19	(8/15-18)	19	19
Aquarium 10 - ♂							
$\bar{X}$	4.257	0.447	0.024	0.308	19	0.0165	72.73
SD	1.3061	0.1371	0.0073	0.0944	(7/28-8/7)	0.0051	22.4939
N	19	19	19	19	18 (8/8-17)	19	19

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray- fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Aug. 18-29							
Aquarium 9 -♀							
$\overline{X}$	2.27	0.238	0.014	0.164	17	0.0096	42.32
SD	0.9402	0.0988	0.0058	0.0680		0.0039	17.2874
N	10	10	10	10		10	10
Aquarium 10 -♂							
$\overline{X}$	1.964	0.200	0.012	0.142	18 (8/18) 17 (8/19-28)	0.0084	37.027
SD	1.2376	0.1271	0.0074	0.0896		0.0050	21.9132
N	10	10	10	10		10	10
Aug. 29-Sept. 9							
Aquarium 9 - ♀							
$\overline{X}$	1.94	0.2037	0.0122	0.1401	17 (8/29-9/6) 16 (9/8)	0.0082	36.15
SD	0.7151	0.0750	0.0045	0.0515		0.0031	13.8726
N	7	7	7	7		7	7
Aquarium 10 -♂							
$\overline{X}$	1.85	0.194	0.013	0.134	17 (8/29) 15 (8/30-9/8)	0,009	39.671
SD	0.7190	0.0755	0.0051	0.0520		0,0036	15,6887
N	7	7	7	7		7	7
Sept. 9-19							
Aquarium 9 -♀							
$\overline{X}$	0.928	0.973	0.0062	0.067	16	0.0042	18.51
SD	0.7416	0.0781	0.0047	0.0540		0.0035	15.5219
N	10	10	10	10		10	10
Aquarium 10 -♂							
$\overline{X}$	0.745	0.078	0.005	0.054	15	0,0036	15,87
SD	0.3899	0.0410	0.0028	0.0282		0.0021	9.1044
N	10	10	10	10		10	10

(continued)

TABLE 21, continued.

	Wet weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray- fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Sept. 20-28							
Aquarium 9 - ♀							
$\bar{X}$	0.910	0.095	0.006	0.0656	16	0.004	17.63
SD	0.5772	0.0606	0.0039	0.0420	(9/20-26)	0.0027	11.9352
N	8	8	8	8	15	8	8
					(9/27-28)		
Aquarium 10 - ♂							
$\bar{X}$	0.668	0.070	0.0046	0.048	15	0.003	13.22
SD	0.2642	0.0277	0.0019	0.0190		0.0013	5.6497
N	8	8	8	8		8	8
Sept. 30-Oct. 4							
Aquarium 9 - ♀							
$\bar{X}$	0.51	0.054	0.0036	0.037	15	0.0024	10.57
SD	0.2009	0.0212	0.0013	0.0147		0.0011	5.0367
N	5	5	5	5		5	5
Aquarium 10 - ♂							
$\bar{X}$	0.492	0.052	0.0036	0.035	15	0.0024	10.57
SD	0.1401	0.0146	0.0009	0.0102		0.0005	2.4100
N	5	5	5	5		5	5
Oct. 5-12							
Aquarium 9 - ♀							
$\bar{X}$	0.503	0.053	0.0037	0.036	15	0,0026	11,46
SD	0.4156	0.435	0.0028	0,0300	(10/5-9)	0,0020	8,7636
N	7	7	7	7	14	7	7
					(10/10-12)		
Aquarium 10 - ♂							
$\bar{X}$	0.55	0.058	0.004	0.040	15	0.003	13.22
SD	0.2489	0.0260	0.0020	0.0179	(10/5-9)	0.0011	4.7105
N	7	7	7	7	14	7	7
					(10/14-12)		

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray-fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Oct. 13-26							
Aquarium 9 - ♀							
$\bar{X}$	0.2485	0.0715	0.0046	0.005	13	0.0	0.0
SD	0.3425	0.0097	0.0008	0.0066		-	-
N	13	13	13	13		13	13
Aquarium 10 - ♂							
$\bar{X}$	0.295	0.031	0.0028	0.021	12	0.0018	7.93
SD	0.2188	0.0231	0.0020	0.0162		0.0012	5.1369
N	13	13	13	13		13	13
Nov. 1-Dec. 2							
Aquarium 9 - ♀							
$\bar{X}$	0.333	0.009	0.001	0.006	11	0.0004	1.76
SD	0.194	0.006	0.0006	0.004	(11/1-8)	0.0005	2.22
N	30	30	30	30	9	30	30
					(11/9-20)		
					8		
					(11/21-26)		
					7		
					(11/27-12/2)		
Aquarium 10 - ♂							
$\bar{X}$	0.327	0.348	0.004	0.024	11	0.003	12.22
SD	0.188	0.020	0.0016	0.014	(11/1-6)	0.001	4.43
N	30	30	30	30	10	30	30
					(11/7-11)		
					9		
					(11/12-18)		
					8		
					(11/19)		
					7		
					(11/20-24)		
					6 (11/25)		
					4		
					(11/26-12/2)		

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray-fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Dec. 3-7							
Aquarium 9 - ♀							
$\bar{X}$	0.180	0.005	0.0006	0.004	7	0.0004	1.76
SD	0.090	0.003	0.0005	0.002		0.0005	2.42
N	5	5	5	5		5	5
Aquarium 10 - ♂							
$\bar{X}$	0.172	0.018	0.004	0.012	4	0.003	12.22
SD	0.048	0.005	0.001	0.004		0.001	4.83
N	5	5	5	5		5	5
Dec. 12-13							
Aquarium 9 - ♀							
$\bar{X}$	0.290	0.008	0.001	0.006	7	0,001	4.41
SD	0.0	0.0	0.0	0.0		0,0	0.0
N	2	2	2	2		2	2
Aquarium 10 - ♂							
$\bar{X}$	0.11	0.021	0.004	0.008	3 (12/12) 1 (12/13)	0.003	12.22
SD	0.0	0.0	0.0	0.0		0.0	0.0
N	2	2	2	2		2	2
Cladophora							
July 20-27							
Aquarium 11 - ♂							
$\bar{X}$	3.44	0.632	0.032	0.563	20	0.028	120.00
SD	1.427	0.263	0.013	0.234		0.012	50.39
N	8	8	8	8		8	8
Aquarium 12 - ♀							
$\bar{X}$	2.67	0.492	0,020	0,438	20	0,022	94,29
SD	1.379	0.254	0,012	0.226		0.011	48.71
N	8	8	8	8		8	8

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray- fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
July 28-Aug. 17							
Aquarium 11 - ♂							
$\bar{X}$	2.07	0.381	0.019	0.340	20	0.017	72.86
SD	1.020	0.188	0.009	0.167		0.0084	36.06
N	20	20	20	20		20	20
Aquarium 12 - ♀							
$\bar{X}$	1.54	0.284	0.015	0.253	20 (7/28-8/10) 19 (8/11-14) 18 (8/15) 17 (8/16-17)	0.013	55.72
SD	0.653	0.120	0.006	0.107		0.0054	23.19
N	20	20	20	20		20	20
Aug. 18-Sept. 8							
Aquarium 11 - ♂							
$\bar{X}$	1.49	0.275	0.016	0.245	20 (8/18-19) 19 (8/19) 17 (8/20-9/5) 16 (9/6-8)	0.014	60.00
SD	0.693	0.127	0.007	0.113		0.006	26.30
N	17	17	17	17		17	17
Aquarium 12 - ♀							
$\bar{X}$	1.72	0.317	0.019	0.282	17	0.017	72.86
SD	0.696	0.128	0.0076	0.114		0.007	28.61
N	17	17	17	17		17	17

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray- fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Sept. 9-19							
Aquarium 11-♂							
$\overline{X}$	0.53	0.097	0.006	0.0865	15	0.006	25.71
SD	0.452	0.083	0.0054	0.074		0.005	22.54
N	10	10	10	10		10	10
Aquarium 12 - ♀							
$\overline{X}$	0.75	0.138	0.008	0.123	17	0.008	34.29
SD	0.659	0.121	0.007	0.108	(9/9-13)	0.007	28.38
N	10	10	10	10	16	10	10
					(9/14-16)		
					15		
Sept. 20-28					(9/18-19)		
Aquarium 11 -♂							
$\overline{X}$	0.44	0.081	0.005	0.072	15	0.0049	21.00
SD	0.380	0.070	0.0046	0.062	(9/20-26)	0.004	18.39
N	8	8	8	8	14	8	8
					(9/27-28)		
Aquarium 12 - ♀							
$\overline{X}$	0.51	0.094	0.006	0.083	15	0.0055	23.57
SD	0.432	0.079	0.005	0.071	(9/20-26)	0.0045	19.44
N	8	8	8	8	14	8	8
					(9/27-28)		
Sept. 30-Oct. 4							
Aquarium 11 - ♂							
$\overline{X}$	0.21	0.038	0.0028	0.034	13	0.0026	11.14
SD	0.163	0.030	0.0025	0.027		0.002	8.89
N	5	5	5	5		5	5
Aquarium 12 - ♀							
$\overline{X}$	0.68	0.125	0.009	0.112	14	0.008	34.29
SD	0.159	0.030	0.002	0.026		0.002	8.02
N	5	5	5	5		5	5

(continued)

TABLE 21, continued.

	Wet Weight gm	Dry Weight gm	Dry gm Consumed/ Crayfish/ day	Ash Free Dry Weight gm	No. Cray- fish	Ash Free Dry Weight/ Crayfish/ day	Cal Consumed/ Crayfish/ day
Oct. 5-11							
Aquarium 11 - ♂							
$\bar{X}$	0.57	0.106	0.0083	0.094	13	0.0074	31.71
SD	0.099	0.018	0.0015	0.016	(10/5-9)	0.0014	5.99
N	7	7	7	7	12	7	7
					(10/10-11)		
Aquarium 12 - ♀							
$\bar{X}$	0.55	0.101	0.007	0.090	14	0.0066	28.28
SD	0.154	0.028	0.002	0.025	(10/5-9)	0.002	7.77
N	7	7	7	7	13	7	7
					(10/10-11)		

* Periphyton - 4302 cal./gm
Cladophora + epiphytes - 4408 cal./gm
Cladophora - 4286 cal./gm

TABLE 22. DATA FOR BASIS OF CONVERSIONS FROM DRY WEIGHT TO CALORIES, 1977 CRAYFISH FOOD EXPERIMENTS

	Dry Weight g	Ash Free Dry Weight g	% Ash Free Dry Weight	Ash Free Dry Weight cal/g
Periphyton				
Dec. 9, 1977	1.020	0.287	28.1	5455
	1.116	0.302	27.1	4382
	1.061	0.296	27.9	4430
	1.053	0.196	18.6	3776
	1.069	0.207	19.4	3832
	1.152	0.245	21.3	3934
$\bar{X}$			23.7	4302
SD			4.5	630
N			6	6
Cladophora + Epiphytes				
Feb. 9, 1978	12.72	9.64	75.8	4480
	13.74	10.18	74.1	4358
	17.94	10.48	58.4	4167
	19.30	10.94	56.7	4406
	12.80	10.40	81.3	4308
	14.98	12.06	80.1	4470
	20.28	13.14	64.8	4379
	19.24	11.50	59.8	4699
$\bar{X}$			68.9	4408
SD			10.1	153.6
N			8	8
Cladophora				
Dec. 9, 1977	0.981	0.870	88.7	4095
	0.960	0.850	88.5	5699
	0.983	0.885	90.0	3972
	0.988	0.872	88.3	4122
	1.004	0.897	89.3	4039
	0.989	0.886	89.6	4038
	0.972	0.865	89.0	4037
$\bar{X}$			89.1	4286
SD			0.6	624.9
N			7	7

TABLE 23. CLADOPHORA BREAKAGE, 1977 EXPERIMENTS[a,b,c]

	Added	Retrieved		Broken		Consumed	
	g	g	%	g	%	g	%
Tray 1 - ♀[d]							
July 23	2.0	0.54	27	0.315	15.8	1.145	57.25
24	2.0	0.70	35	0.137	6.9	1.163	58
25	2.0	0.27	13.5	0.28	14	1.45	72.5
26	2.0	0.015	0.75	0.41	20.5	1.575	79
27	2.0	0.29	14.5	0.238	11.9	1.472	73.6
$\overline{X}$		0.36	18.2	0.28	13.8	1.36	68.1
SD		0.26	13.2	0.10	5.0	0.19	9.9
N		5	5	5	5	5	5
Tray 2 - ♂[d]							
July 23	2.0	0.23	11.5	0.41	21	1.36	68
24	2.0	0.64	32	0.27	13.5	1.09	54.5
25	2.0	0.82	41	0.34	17	0.84	42
26	2.0	0.63	31.5	0.165	8.25	1.205	60.3
27	2.0	0.53	26.5	0.305	15.3	1.165	58
$\overline{X}$		0.57	28.5	0.30	15.0	1.13	56.6
SD		0.22	10.9	0.09	4.7	0.19	9.5
N		5	5	5	5	5	5

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[a] Retrieved: that portion easily removable with forceps.
Broken: that portion of very small filaments found on the bottom of the tray and collected by filtration.
Consumed: calculated by subtracting the retrieved and broken portions from the amount added.

[b] All weights are wet weights.

[c] Four crayfish were in each tray, except Trays 3 and 6, which were empty.

[d] Food in for 16 hours each day.

(continued)

TABLE 23, continued.

	Added	Retrieved		Broken		Consumed	
	g	g	%	g	%	g	%
Tray 3 - empty[d]							
July 23	2.0	2.0	100	0.015	0.75	0	0
24	2.0	1.87	93.5	0.02	1.0	0.11	6.5
25	2.0	1.83	91.5	0.025	1.3	0.145	7.3
26	2.0	1.75	87.5	0.045	2.3	0.205	12.5
27	2.0	2.0	100	0.05	2.5	0	0
$\overline{X}$		1.89	94.5	0.03	1.57	0.09	5.3
SD		0.11	5.5	0.02	0.79	0.09	5.3
N		5	5	5	5	5	5
Tray 4 - ♀[e]							
July 25	2.0	0.95	47.5	0.276	13.8	0.774	38.7
26	2.0	0.76	38	0.312	15.6	0.928	46.4
27	2.0	0.53	26.5	0.320	16.0	1.15	57.5
28	2.0	0.75	37.5	0.100	5.0	1.15	57.5
29	2.0	0.78	39	0.275	13.8	0.945	47.3
$\overline{X}$		0.75	37.7	0.26	12.8	0.99	49.5
SD		0.15	7.5	0.09	4.5	0.16	8.0
N		5	5	5	5	5	5
Tray 5 - ♂[e]							
July 25	2.0	1.23	61.5	0.262	13.1	0.508	25.4
26	2.0	0.36	43	0.338	16.9	0.802	40
27	2.0	1.08	54	0.28	14.0	0.64	32
28	2.0	1.06	53	0.21	10.5	0.73	36.5
29	2.0	0.90	45	0.285	14.3	0.815	40.8
$\overline{X}$		0.93	51.3	0.28	13.8	0.70	34.9
SD		0.34	7.5	0.05	2.5	0.13	6.4
N		5	5	5	5	5	5

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[e] Food in for 4 hours each day.

(continued)

TABLE 23, continued.

	Added	Retrieved		Broken		Consumed	
	g	g	%	g	%	g	%
Tray 6 - empty[e]							
July 25	2.0	1.725	86.3	0.075	3.8	0.2	10
26	2.0	1.79	89.5	0.021	1.1	0.189	9.5
27	2.0	1.82	91.0	0.115	5.8	0.065	3
28	2.0	1.74	87.0	0.065	3.3	0.195	9.8
29	2.0	1.83	91.5	0.065	3.3	0.105	5
$\overline{X}$		1.78	89.1	0.070	3.5	0.15	7.5
SD		0.05	2.3	0.03	1.7	0.06	3.2
N		5	5	5	5	5	5

TABLE 24. CRAYFISH FECES, 1977 EXPERIMENTS

	Dry Weight (mg)		No. Crayfish		Dry Weight of Feces/Crayfish	
	Aquar. 7 (♀)	Aquar. 8 (♂)	Aquar. 7 (♀)	Aquar. 8 (♂)	Aquar. 7 (♀)	Aquar. 8 (♂)
Periphyton						
Aug. 15	31.2	19.5	17	13	1.84	1.50
23	23.1	38.7	14	13	1.65	2.98
25	50.4	42.3	13	13	3.88	3.25
30	24.2	41.3	13	11	1.86	3.75
Sept. 6		6.6		10		0.66
16	15.9	27.5	12	10	1.33	2.75
21	1.6	22.6	12	9	0.13	2.51
22	1.3	14.7	12	9	0.11	1.63
23	12.8	27.0	12	9	1.07	3.00
27	15.6	3.4	12	9	1.30	0.38
Oct. 26	10.7	0.8	9	6	1.19	0.13
27	16.9	5.2	9	6	1.88	0.87
28	6.6	4.9	9	6	0.73	0.82
Nov. 9	4.4	0.5	9	6	0.49	0.08
11	1.8	0.8	9	6	0.20	0.01
$\overline{X}$	15.46	17.05			1.26	1.62
SD	13.63	15.42			0.99	1.31
N	14	15			14	15

(continued)

TABLE 24, continued.

	Dry Weight (mg)		No. Crayfish		Dry Weight of Feces/Crayfish	
	Aquar. 9 (♀)	Aquar. 10 (♂)	Aquar. 9 (♀)	Aquar. 10 (♂)	Aquar. 9 (♀)	Aquar. 10 (♂)
Cladophora + Epiphytes						
Aug. 15	32.0		18		1.78	
23	14.2	23.1	17	17	0.84	1.36
25	17.1	5.6	17	17	1.01	0.33
30	4.2	8.4	17	15	0.25	0.56
Sept. 6	6.5	3.0	17	15	0.38	0.20
16	3.9	2.8	16	15	0.24	0.19
21	3.0	0.7	16	15	0.19	0.05
22	6.9	0.5	16	15	0.43	0.03
23	1.8	0.8	16	15	0.11	0.05
27	1.3	0.4	15	15	0.09	0.03
Oct. 7	3.7	4.6	15	15	0.25	0.31
$\overline{X}$	8.60	4.99			0.51	0.31
SD	9.25	6.88			0.51	0.41
N	11	10			11	10
	Aquar. 12 (♀)	Aquar. 11 (♂)	Aquar. 12 (♀)	Aquar. 11 (♂)	Aquar. 12 (♀)	Aquar. 11 (♂)
Cladophora						
Aug. 23	4.7	4.6	17	17	0.28	0.27
25	14.8	1.0	17	17	0.87	0.06
30	4.7	3.9	17	17	0.28	0.23
Sept. 6		2.5		16		0.16
16	0.1	0.5	16	15	0.01	0.03
21	0.0	0.2	15	15	0.0	0.01
22	0.2	0.2	15	15	0.01	0.01
23	3.3	3.8	15	15	0.22	0.25
27		0.8		14		0.06
$\overline{X}$	3.97	1.94			0.24	0.12
SD	5.22	1.77			0.31	0.11
N	7	9			7	9

TABLE 25. UTILIZATION OF CALORIES PER CRAYFISH, 1977 CRAYFISH EXPERIMENT

	Tissue* $\bar{X}$ Cal./ Crayfish	Gain or Loss	Resp. $\bar{X}$ Cal./ Crayfish	Total Cal. Assim.	% Wt. Gain or Loss	% Resp.	Total Food Ingest.	% Wt. Gain or Loss	% Resp.	% Ingest. Assim.
Periphyton										
July 18 (Start of Expt.)										
♀	4288									
♂	5016									
Oct. 3-4										
♀	4639	351	6301.68	6652.68	5.3	94.7	207,314	0.2	3.04	3.2
♂	5686	670	6116.88	6786.88	4.9	90.1	136,074	0.5	4.50	5.0
Dec. 13-14										
♀	4563	- 73	1168	1741	4.2	95.8				
♂	5249	-437	928.8	1365.8	32.0	68.0				
Cladophora + Epiphytes										
July 18										
♀	3011									
♂	3995									
Oct. 3-4										
♀	3034	23	3152.88	3175.88	0.7	99.3	3737.13	0.62	84.4	85
♂	3823	(-172)	3215.52	3387.52	5.1	94.9	3565.62	2.0	90.2	95
Dec. 13-14										
♀	2821	-916.13	610.1	1526.4	(60)	40	194.2	471.8	314.1	786
♂	2829	-736.62	491.0	1227.62	(60)	40	810.31	90.9	60.6	151.5

(continued)

TABLE 25, continued.

	Tissue $\bar{X}$ Cal./ Crayfish	Gain or Loss	Resp. $\bar{X}$ Cal./ Crayfish	Total Cal. Assim.	% Wt. Gain or Loss	% Resp.	Total Food Ingest.	% Wt. Gain or Loss	% Resp.	% Ingest. Assim.
Cladophora										
July 18										
♀	2800									
♂	3068									
Oct. 3-4										
♀	2918	118	2698	2816	4.2	95.8	4170.5	2.8	64.7	67.5
♂	3170	102	4158	4260	2.4	97.6	4286	2.4	97.01	99.4
Dec. 13-14										
♀	2838	- 80	1289	1369	(5.8)	94.2				
♂	2854	-316	882	1198	(26.4)	63.6				

* Periphyton - 4498.8 cal./gm - ♀
3897 cal./mg - ♂
Cladophora + epiphytes - 3922.49 cal./gm - ♀
3703.18 cal./gm - ♂
Cladophora - 3761 cal./mg - ♀
3250 cal./mg - ♂

TABLE 26. CRAYFISH MOLTING EXPERIMENT, 1977-78

	Cray-fish Desig.	Wet Wt.(g) Start	Wet Wt.(g) End	Dry Wt.(g) Calcu.	% Dry Wt.(g)	Molted	Dry Wt.(g) End	% Gain or Loss Wet Wt.	% Gain or Loss Dry Wt.	No. of Days Alive
Periphyton Aquarium 7 ♂	R-B	4.52	5.10	0.65	14.4	yes	0.735	+11.4	+11.6	18 (12/14-12/31)
	B	2.90	3.28	0.47	16.1	yes	0.541	+11.6	+13.1	25 (12/14-1/7)
	R-R	4.10	4.47	0.68	16.5	yes	0.739	+ 8.3	+ 8.0	20 (12/14-1/2)
	Br	4.56	4.89	0.69	15.1	yes	0.736	+ 6.7	+ 6.3	21 (12/14-1/3
Aquarium 8 ♀	R-B	6.30	5.52	1.76	27.9	partial	1.54	-12.4	-12.5	14 (12/14-12/27)
	Br	7.25	7.04	1.99	27.4	no	1.93	- 2.9	- 3.0	15 (12/14-12/28)
	Br-R-B	3.90	3.81	1.04	26.8	no	1.02	- 2.3	- 1.9	14 (12/14-12/27)
Cladophora + Epiphytes Aquarium 9 ♂	Br-B	4.57	4.44			no		- 2.8		15 (12/14-12/28)
	R-B	4.90	4.73			no		- 3.5		20 (12/14-1/2)
Cladophora Aquarium 12 ♀	Br-R-B	4.07	4.08	0.96	23.6	no	0.963	even	even	14 (12/14-12/27)
	B-Br	7.00	7.24	1.74	24.9	no	1.80	+ 3.3	+ 3.3	13 (12/14-12/26)
	B	3.76	3.63	0.88	23.5	no	0.857	- 3.5	- 3.2	18 (12/14-12/31

TABLE 27. INDIVIDUAL TISSUE WEIGHTS, 1977 TADPOLE EXPERIMENTS

	Wet Weight mg/tadpole		Dry Weight mg/tadpole	
Start of Experiment April 16	9.0	10.3	0.79	0.92
	10.0	10.4	0.82	0.73
	13.5	9.6	1.00	0.73
	12.7	9.4	0.10	0.82
	15.0	10.6	0.11	0.85
	11.0	11.1	0.80	0.89
	12.2	11.3	0.95	0.77
	12.3	9.9	0.92	0.84
	12.1	11.7	0.89	0.84
	10.6	11.2	0.10	0.86
	10.5	13.4	0.75	0.88
	12.7	11.1	0.89	0.96
	11.4	8.0	0.91	0.14
	11.6	8.4	0.87	0.29
	10.4	13.7	0.91	0.73
	11.4	6.3	0.79	0.42
	10.6	11.4	0.91	0.85
	8.3	12.8	0.81	0.93
	10.1	10.9	0.73	0.78
	9.9	10.3	0.78	0.85
$\bar{X}$	10.93		0.748	
SD	1.68		0.250	
N	40		40	

	Wet Weight mg/tadpole	Dry Weight mg/tadpole		Wet Weight mg/tadpole	Wet Weight mg/tadpole
Periphyton April 22	351.5	39.48	May 5	1252	130.5
	382.1	31.03		915	167.7
	234.7	30.96		819	181.5
	320.0	20.56		691	123.1
	238.1	20.74		907	126.3
	273.6	28.17		1072	183.7
	202.7	22.59		893	101.5
	203.5			1390	79.5
$\bar{X}$	275.8	27.65		992	136.7
SD	68.3	6.91		232	37.8
N	8	7		8	8

(continued)

TABLE 27, continued.

	Wet Weight mg/tadpole	Dry Weight mg/tadpole		Wet Weight mg/tadpole	Dry Weight mg/tadpole
Periphyton					
May 18	2570	436	June 1	2430	915
	2670	459		3690	563
	2340	381		5370	913
	2820	407		5450	692
	2950	464		4130	333
	2510	331		5270	757
	2520	358		4960	786
	3340	584		5310	764
	2470	351		2920	722
	2980	432		4840	423
$\bar{X}$	2717	420		4437	687
SD	301	74		1094	190
N	10	10		10	10
Cladophora + Epiphytes					
April 22	302.9	31.97	May 5	1206	113.6
	301.5	28.74		1266	183.3
	277.9	32.39		1335	184.8
	400.3	21.78		768	215.8
	285.4	38.30		944	189.1
	361.7	29.21		1547	164.5
	447.1	47.55		1104	138.1
	447.5	48.55		1144	247.8
$\bar{X}$	356.8	34.81		1164	179.6
SD	77.4	9.37		238	42.1
N	8	8		8	8
May 18	2140	641	June 1	4930	712
	3720	303		4690	509
	2490	328		3710	731
	3920	585		5740	842
	3200	525		6510	1009
	2730	235		2930	668
	3010	434		3960	437
	1540	393		6380	899
	2910	425		5330	1176
	3090	376		7200	1114
$\bar{X}$	2875	425		5138	810
SD	705	127		1359	245
N	10	10		10	10

(continued)

TABLE 27, continued.

	Wet Weight mg/tadpole	Dry Weight mg/tadpole		Wet Weight mg/tadpole	Dry Weight mg/tadpole
Cladophora					
April 22	9.8	0.580	May 6	12.0	0.458
	9.5	0.404		9.3	0.700
	7.7	0.430		10.6	0.842
	10.0	0.484		13.8	0.516
	10.7	0.564		19.5	0.382
	11.7	0.440		11.2	0.382
	10.3	0.596		7.3	0.454
	9.9	0.574		7.8	0.328
	12.4	0.510		12.5	0.332
	9.3	0.510		14.4	0.550
				22.8	1.154
				31.3	0.826
$\bar{X}$	10.1	0.509		14.4	0.547
SD	1.3	0.069		7.0	0.241
N	10	10		12	12
May 18	83.5	0.938			
	102.0	1.472			
	20.9	0.772			
	18.7	0.634			
	18.5	0.666			
	34.3	0.224			
	19.7	0.402			
	29.6	0.290			
	28.0	1.518			
	13.7	0.528			
	27.6	0.820			
$\bar{X}$	36.0	0.751			
SD	29.0	0.428			
N	11	11			

TABLE 28. RESPIRATION, 1977 TADPOLE EXPERIMENTS

	Dry Weight (mg)			ul O_2/mg/hr				cal/tadpole/	cal/tadpole/
	$\bar{X}$	SD	N	$\bar{X}$	SD	N	cal/mg/hr*	hr	day
Periphyton									
April 5	136.7	37.8	8	0.89	0.09	4	0.0042	0.574	13.78
April 18	420.0	74.0	10	0.35	0.10	3	0.0016	0.714	17.14
Cladophora + Epiphytes									
April 5	179.6	42.1	8	0.89	0.36	4	0.0042	0.754	18.10
April 18	424.0	128.0	10	0.65	0.15	4	0.0031	1.300	31.09
Cladophora									
April 5	0.547	0.241	12	2.41	0.88	4	0.0114	0.006	0.149
April 18	0.751	0.430	11	3.46	1.23	4	0.0163	0.012	0.293

* 1 ul O_2 = 0.00472 calories.

TABLE 29. TISSUE WEIGHTS, 1977 CATFISH EXPERIMENTS

	Wet Weights, mg/fish							
	Tank 1		Tank 2		Tank 5		Tank 6	
Start of Experiment June 17	470	420	480	490	660	530	330	490
	750	520	330	520	460	450	590	550
	460	490	410	480	440	340	510	500
	580	510	550	460	370	460	330	540
	530	580	510	630	700	540	320	440
	510	460	580	540	520	510	320	660
	550	400	390	580	530	460	340	590
	590	570	400	490	410	460	500	410
	400	540	430	310	400	500	580	520
	510	450	410	480	560	400	490	370
$\bar{X}$	515		474		485		469	
SD	80		83		89		105	
N	20		20		20		20	

Periphyton

	Tank 5		Tank 6			Tank 5		Tank 6	
July 1	540	690	580	540	July 8	850	780	1580	950
	560	550	670	710		530	1140	910	560
	480	660	760	570		1060	860	1180	740
	440	540	430	440		900	710	1280	930
	530	450	350	570		990	1320	920	770
	440	500	780	740		1490	670	650	1410
	470		770	540		810	1390		
			500	570		480	980		
			500	610					
$\bar{X}$	527		596			940		990	
SD	78		124			290		310	
N	13		18			16		12	

	Tank 5		Tank 6	
July 15	720	470	430	580
	460	570	310	590
	520	550	360	550
	460	390	540	610
	470	540	570	710
	510	490	810	540
	460	410	530	740
	490	570	500	400
	460	350	710	330
			580	
$\bar{X}$	490		550	
SD	80		140	
N	18		19	

(continued)

TABLE 29, continued

	Wet Weights, mg/fish								
	Tank 5		Tank 6			Tank 5		Tank 6	
Cladophora									
July 1	360	380	330	260	July 8	260	260	200	330
	370	370	460	250		450	250	210	250
	470	410	190	490		280	330	190	210
	380	460	350	270		230	280	280	280
	370	360	390	340		250	260	290	300
	320	320	360	350		270	270	280	290
	350	370	360	310		190	250	260	330
	370	310	190	320		270	390	310	
	550	320	390	390		340			
			330						
$\overline{X}$	380		333			280		270	
SD	60		78			60		50	
N	18		19			17		15	

TABLE 30. FATHEAD MINNOW TISSUE WEIGHTS, 1978 EXPERIMENT

	Wet wt. gm/fish		Dry wt.[a] gm/fish		AFDW[b] gm/fish		Cal./fish AFDW[c]	
Start of Experiment								
Periphyton	1.14	1.36	0.301	0.359	0.269	0.321	1716	2047
March 7	0.95	1.07	0.132	0.282	0.118	0.252	0753	1607
Tray 1	0.85	1.25	0.224	0.330	0.200	0.295	1276	1882
	1.22	1.03	0.322	0.272	0.288	0.243	1837	1550
	0.92	1.16	0.243	0.306	0.217	0.273	1384	1741
	0.77	0.94	0.203	0.248	0.181	0.221	1154	1410
	1.77	1.44	0.467	0.380	0.417	0.339	2660	2162
	1.70	0.95	0.449	0.132	0.401	0.118	2558	0753
	1.01	1.10	0.267	0.290	0.238	0.259	1518	1652
	1.56	1.29	0.412	0.341	0.368	0.305	2347	1945
$\overline{X}$	1.17		0.298		0.266		1700	
SD	0.28		0.090		0.081		520	
N	20		20		20		20	

[a] All wet to dry conversions for Start of Experiment, March 7, based on 26.4%

[b] 89.3% of dry weight, March 3, see Table 21.

[c] Based on 6378 cal/gm, March 3, see Table 21.

Tray 2	0.73	0.62	0.193	0.164[a]	0.172	0.146[b]	1097	0931[c]
	0.92	0.90	0.243	0.238	0.217	0.213	1384	1359
	0.50	0.56	0.132	0.148	0.118	0.132	0753	0842
	0.80	0.75	0.211	0.199	0.188	0.178	1199	1135
	0.95	0.72	0.251	0.190	0.224	0.170	1429	1084
	0.67	0.84	0.177	0.222	0.158	0.198	1008	1263
	0.75	0.72	0.199	0.190	0.178	0.170	1135	1084
	0.94	0.96	0.248	0.253	0.221	0.226	1410	1441
	0.67	0.71	0.177	0.187	0.158	0.167	1008	1065
	0.79	0.75	0.209	0.199	0.187	0.178	1193	1135
$\overline{X}$	0.76		0.202		0.180		1148	
SD	0.13		0.034		0.030		193	
N	20		20		20		20	

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt.[a] gm/fish		AFDW[b] gm/fish		Cal./fish AFDW[c]	
Start of Experiment								
Cladophora + epiphytes								
March 7	0.83	0.76	0.219	0.201	0.196	0.179	1250	1142
Tray 3	0.63	0.56	0.166	0.148	0.148	0.132	0944	0842
	0.78	0.74	0.206	0.195	0.184	0.174	1174	1110
	0.65	0.72	0.172	0.190	0.154	0.170	0982	1084
	0.90	0.63	0.238	0.166	0.213	0.148	1359	0944
	0.71	0.84	0.187	0.222	0.167	0.198	1065	1263
	0.95	1.11	0.251	0.293	0.224	0.262	1429	1671
	0.71	0.76	0.187	0.201	0.167	0.179	1065	1142
	0.99	0.73	0.261	0.193	0.233	0.172	1486	1097
	0.71	0.77	0.187	0.203	0.167	0.181	1065	1154
$\overline{X}$	0.78		0.204		0.182		1163	
SD	0.140		0.035		0.031		201	
N	20		20		20		20	
Tray 4	1.15	0.96	0.304	0.253[a]	0.271	0.226[b]	1728	1441[c]
	0.90	1.18	0.238	0.312	0.212	0.279	1352	1779
	1.05	1.07	0.277	0.282	0.247	0.252	1575	1607
	1.30	1.08	0.343	0.285	0.306	0.255	1952	1626
	1.07	1.15	0.282	0.304	0.252	0.271	1607	1728
	1.09	1.09	0.288	0.288	0.257	0.257	1639	1639
	1.12	1.00	0.296	0.264	0.264	0.236	1684	1505
	1.06	1.44	0.280	0.380	0.250	0.339	1595	2162
	1.14	1.04	0.301	0.275	0.269	0.246	1716	1569
	1.49	1.15	0.393	0.304	0.351	0.271	2239	1728
$\overline{X}$	1.13		0.297		0.266		1694	
SD	0.14		0.038		0.034		214	
N	20		20		20		20	
Start of Experiment								
Cladophora	0.66	0.74	0.174	0.195[a]	0.155	0.174[b]	989	1110[c]
March 9	0.63	0.95	0.166	0.251	0.148	0.224	944	1429
Tray 5	0.97	1.13	0.256	0.298	0.229	0.266	1461	1697
	0.84	0.71	0.222	0.187	0.198	0.167	1263	1065
	0.68	0.98	0.180	0.259	0.161	0.231	1027	1473
	0.98	0.76	0.259	0.201	0.231	0.179	1473	1142
	1.04	0.79	0.275	0.209	0.246	0.187	1569	1193
	0.70	0.55	0.185	0.145	0.165	0.129	1052	823
	0.77	0.98	0.203	0.259	0.181	0.231	1154	1473
	0.90	0.91	0.238	0.240	0.212	0.214	1352	1365
$\overline{X}$	0.83		0.220		0.196		1253	
SD	0.16		0.041		0.037		236	
N	20		20		20		20	

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt. [a] gm/fish		AFDW [b] gm/fish		Cal./fish AFDW [c]	
Tray 6								
	0.64	1.04	0.169	0.275	0.151	0.246	963	1569
	1.23	0.92	0.325	0.243	0.290	0.217	1850	1384
	1.30	0.88	0.343	0.232	0.306	0.207	1952	1320
	0.87	1.18	0.230	0.312	0.205	0.279	1307	1779
	1.02	1.22	0.269	0.322	0.240	0.288	1531	1837
	1.13	1.04	0.298	0.275	0.266	0.246	1697	1569
	0.84	1.42	0.222	0.375	0.198	0.335	1263	2137
	0.93	0.91	0.246	0.240	0.220	0.214	1403	1365
	0.84	0.80	0.222	0.211	0.198	0.188	1263	1199
	1.00	1.03	0.264	0.272	0.236	0.243	1505	1550
$\overline{X}$	1.01		0.267		0.239		1522	
SD	0.19		0.050		0.045		287	
N	20		20		20		20	
Periphyton								
March 29	1.34	1.29	0.323	0.311[d]	0.286	0.275[e]	1735	1668[f]
Tray 1	1.05	1.88	0.253	0.453	0.224	0.401	1359	2432
	1.57	1.42	0.378	0.342	0.335	0.303	2032	1838
	1.44	1.37	0.347	0.330	0.307	0.292	1862	1771
	1.92	1.22	0.463	0.294	0.410	0.260	2487	1577
	2.37	1.20	0.571	0.289	0.505	0.256	3063	1553
	1.13	1.17	0.272	0.282	0.241	0.250	1462	1516
	1.59	1.44	0.383	0.347	0.339	0.307	2056	1862
$\overline{X}$	1.46		0.352		0.312		1892	
SD	0.34		0.083		0.074		446	
N	16		16		16		16	

[d] 24.1% of wet weight based on April 8-11 values.
[e] 88.5% of dry weight, based on April 8-11 values, see Table 21.
[f] 6065 cal/gm based on April 8-11 values, see Table 21.

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt.[g] gm/fish		AFDW [e] gm/fish		Cal./fish AFDW[f]	
March 29	1.04	0.95	0.251	0.229	0.222	0.203	1346	1231
Tray 2	0.79	1.02	0.190	0.246	0.168	0.218	1019	1322
	1.03	1.35	0.248	0.326	0.219	0.289	1328	1753
	1.19	1.05	0.287	0.253	0.254	0.224	1541	1359
	1.19	1.27	0.287	0.306	0.254	0.271	1541	1644
	0.78	0.89	0.188	0.214	0.166	0.189	1007	1146
	1.63	1.19	0.393	0.287	0.348	0.254	2111	1541
	0.71		0.171		0.151		916	
$\overline{X}$	1.07		0.258		0.229		1387	
SD	0.24		0.058		0.052		315	
N	15		15		15		15	

[g] 24.2% of wet weight based on April 8-11 values.

April 14	1.27	1.27	0.306	0.306[d]	0.271	0.271[e]	1644	1644[f]
Tray 1	2.02	1.31	0.487	0.316	0.431	0.280	2614	1698
	1.63	1.53	0.393	0.369	0.348	0.327	2111	1983
	1.89		0.455		0.403		2444	
$\overline{X}$	1.560		0.376		0.333		2020	
SD	0.305		0.073		0.065		393	
	7		7		7		7	
April 14								
Tray 2	0.83	0.80	0.201	0.194[g]	0.178	0.172[e]	1080	1043[f]
	1.08	1.02	0.261	0.247	0.231	0.219	1401	1328
	1.20	1.23	0.290	0.298	0.257	0.264	1559	1601
	0.80	1.20	0.194	0.290	0.172	0.257	1043	1559
$\overline{X}$	1.020		0.247		0.219		1327	
SD	0.187		0.045		0.040		242	
N	8		8		8		8	

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt.[g] gm/fish		AFDW [h] gm/fish		Cal./fish AFDW[i]	
May 16-23	1.95	1.34	0.513	0.352	0.458	0.314	2886	1979
Tray 1	2.05	1.34	0.539	0.352	0.481	0.314	3031	1979
X̄	1.670		0.439		0.392		2469	
SD	0.383		0.101		0.090		569	
	4		4		4		4	

[h] 26.3% of wet weight, based on May 16-20 values.
[i] 89.2% of wet weight, based on May 16-20 values.

May 16-23								
Tray 2	1.10	1.11	0.272	0.274[k]	0.243	0.244[i]	1531	1537[j]
	1.32	0.85	0.326	0.210	0.291	0.187	1834	1178
X̄	1.095		0.271		0.242		1520	
SD	0.192		0.047		0.041		268	
N	4		4		4		4	

[j] 6301 cal/gm, May 16-20, see Table 21.
[k] 24.7% of wet weight, based on May 16-20 values, Table 21.

Cladophora & Epiphytes								
March 29	0.73	0.65	0.151	0.135[l]	0.125	0.112[m]	736	659[n]
Tray 3	0.50	0.61	0.104	0.126	0.086	0.105	506	618
	0.63	0.58	0.130	0.120	0.108	0.100	636	589
	0.55	0.75	0.114	0.155	0.095	0.129	559	759
	0.75	0.99	0.155	0.205	0.129	0.170	759	1000
	0.85	0.67	0.176	0.139	0.146	0.115	859	677
	0.58	0.62	0.120	0.128	0.100	0.106	589	624
	0.49	0.68	0.101	0.141	0.084	0.117	494	689
X̄	0.66		0.140		0.114		672	
SD	0.13		0.030		0.022		130	
N	16		16		16		16	

[l] 20.7% of wet weight, based on April 8-11 values.
[m] 83% of dry weight, based on April 8-11 values, see Table 21.
[n] 5885 cal/gm based on April 8-11 values, see Table 21.

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt.[o] gm/fish		AFDW [m] gm/fish		Cal./fish AFDW [n]	
March 29								
Tray 4	1.00	0.87	0.199	0.173	0.165	0.144	971	847
	0.72	0.87	0.143	0.173	0.119	0.144	700	847
	0.99	0.94	0.197	0.187	0.164	0.155	965	912
	1.12	0.88	0.223	0.175	0.185	0.145	1089	853
	0.91	0.83	0.181	0.165	0.150	0.137	883	806
	0.89	1.00	0.177	0.199	0.147	0.165	865	971
	1.33	0.87	0.265	0.173	0.220	0.144	1295	847
	1.30	0.89	0.259	0.177	0.215	0.147	1265	865
$\overline{X}$	0.96		0.192		0.159		936	
SD	0.16		0.033		0.027		159	
N	16		16		16		16	

[o] 19.9% of wet weight, based on April 8-11 values.

	Wet wt. gm/fish		Dry wt.[o] gm/fish		AFDW [m] gm/fish		Cal./fish AFDW [n]	
April 14	0.47	0.53	0.097	0.110[l]	0.081	0.091[m]	477	536[n]
Tray 3	0.59	0.68	0.122	0.141	0.101	0.119	594	700
	0.49	0.57	0.101	0.118	0.084	0.098	494	577
	0.45	0.59	0.095	0.122	0.079	0.101	465	594
	0.58		0.100		0.083		488	
$\overline{X}$	0.550		0.112		0.093		547	
SD	0.072		0.015		0.013		76	
N	9		9		9		9	
April 14								
Tray 4	0.98	0.86	0.195	0.171[o]	0.162	0.142[m]	953	836[n]
	0.62	0.82	0.123	0.163	0.102	0.135	600	794
	1.01	0.76	0.201	0.151	0.167	0.125	983	736
	1.22	0.92	0.243	0.183	0.202	0.152	1189	895
	0.83		0.165		0.137		806	
$\overline{X}$	0.891		0.177		0.147		866	
SD	0.170		0.034		0.028		167	
N	9		9		9		9	

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt.[p] gm/fish		AFDW[q] gm/fish		Cal./fish AFDW[r]	
May 16-23								
Tray 3	0.38	0.54	0.051	0.072	0.039	0.055	201	284
	0.66	0.62	0.088	0.082	0.067	0.062	346	320
$\overline{X}$	0.550		.073		0.056		288	
SD	0.124		0.016		0.012		63	
	4		4		4		4	

[p] 14.9% of wet weight, based on May 16-20 values.
[q] 76% of dry weight, based on May 16-20 values, see Table 21.
[r] 5165 cal/gm based on May 16-20 values, see Table 21.

	Wet wt. gm/fish		Dry wt. gm/fish		AFDW gm/fish		Cal./fish AFDW	
Tray 4	0.88	1.06	0.131	0.158[p]	0.100	0.120[q]	517	620[r]
	0.62	0.77	0.092	0.115	0.070	0.087	362	449
$\overline{X}$	0.833		0.124		0.094		487	
SD	0.185		0.028		0.021		109	
N	4		4		4		4	
Cladophora								
March 29								
Tray 5	0.49	1.01	0.098	0.203[s]	0.084	0.175[t]	515	1073[u]
	0.71	0.62	0.143	0.125	0.123	0.108	754	662
	0.59	0.73	0.119	0.147	0.103	0.127	632	779
	0.54	0.92	0.109	0.185	0.094	0.159	576	975
	0.78	0.51	0.157	0.103	0.135	0.089	828	546
	0.89	0.76	0.179	0.153	0.154	0.132	944	810
	0.61	0.65	0.123	0.131	0.106	0.113	650	693
	0.78	0.79	0.157	0.159	0.135	0.137	828	840
$\overline{X}$	0.71		0.143		0.123		757	
SD	0.15		0.030		0.026		159	
N	16		16		16		16	

[s] 20.1% of wet weight, based on April 8-11 values.
[t] 86.2% of dry weight, based on April 8-11 values, see Table 21.
[u] 6133 cal/gm based on April 8-11 values, see Table 21.

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt.[v] gm/fish		AFDW[t] gm/fish		Cal./fish/AFDW[u]	
March 29 Tray 6	0.58	0.80	0.124	0.170	0.107	0.147	656	902
	0.98	1.05	0.209	0.224	0.180	0.193	1104	1184
	0.93	1.04	0.198	0.222	0.171	0.191	1049	1171
	0.93	0.86	0.198	0.183	0.171	0.158	1049	969
	0.73	1.16	0.155	0.247	0.134	0.213	822	1306
	0.81	0.78	0.173	0.165	0.149	0.142	914	871
	0.70	0.69	0.149	0.147	0.128	0.127	785	779
	0.85		0.181		0.156		957	
$\bar{X}$	0.86		0.183		0.158		968	
SD	0.16		0.034		0.029		177	
N	15		15		15		15	

[v] 21.3% of wet weight, based on April 8-11 values.

	Wet wt. gm/fish		Dry wt. gm/fish		AFDW gm/fish		Cal./fish/AFDW	
April 14 Tray 5	0.52	0.82	0.105	0.165[s]	0.091	0.142[t]	558	871[u]
	0.69	0.45	0.139	0.090	0.120	0.078	736	478
	0.72	0.72	0.145	0.145	0.125	0.125	767	767
	0.90	0.64	0.181	0.129	0.156	0.111	957	681
	0.54		0.109		0.094		577	
$\bar{X}$	0.667		0.134		0.116		710	
SD	0.146		0.029		0.025		154	
N	9		9		9		9	
Tray 6	0.84	0.81	0.179	0.173[v]	0.154	0.149[t]	944	914[u]
	0.85	1.06	0.181	0.226	0.156	0.195	957	1196
	0.73	0.73	0.155	0.155	0.134	0.116	822	711
$\bar{X}$	0.837		0.178				924	
SD	0.121		0.261				162	
N	6		6				6	

(continued)

TABLE 30, continued.

	Wet wt. gm/fish		Dry wt.[w] gm/fish		AFDW[x] gm/fish		Cal./fish/AFDW[y]	
May 16-23								
Tray 5	0.55	0.65	0.082	0.097	0.062	0.074	325	389
	0.86	0.32	0.128	0.048	0.097	0.037	509	194
	0.39		0.058		0.044		231	
$\overline{X}$	0.554		0.083		0.063		330	
SD	0.215		0.032		0.024		126	
N	5		5		5		5	

[w] 14.1% of wet weight, based on May 16-23 values.
[x] 76% of dry weight, based on May 16-23 values, see Table 21.
[y] 5244 cal/gm, based on May 16-23 values, see Table 21.

Tray 6	0.74	0.79	0.104	0.111[w]	0.079	0.084[x]	414	440[y]
	0.78	1.02	0.120	0.144	0.091	0.110	477	577
	0.63	0.66	0.089	0.093	0.068	0.071	357	372
$\overline{X}$	0.770		0.110		0.084		440	
SD	0.138		0.020		0.153		80	
N	6		6		6		6	

TABLE 31. DATA FOR BASIS OF CONVERSIONS FROM DRY WEIGHT TO CALORIES, 1978 FATHEAD MINNOW TISSUE EXPERIMENTS

	Dry Weight (g)	AFDW (g)	% AFDW	cal/g
March 3 Start of Experiment	0.4712	0.4205	89.2	6,404
	0.5355	0.4842	90.4	6,431
	0.5539	0.4952	89.4	6,320
	0.4440	0.3869	87.1	6,297
	0.5114	0.4614	90.2	6,437
$\bar{X}$			89.3	6,378
SD			1.31	65
N			5	5
April 8-11 *Cladophora*	0.4420	0.3804	86.0	6,174
	0.4410	0.3806	86.4	6,091
$\bar{X}$			86.2	6,133
SD			0.28	59
N			2	2
Cladophora + Epiphytes	0.4203	0.3511	83.5	6,103
	0.2847	0.2349	82.5	5,666
$\bar{X}$			83.0	5,885
SD			0.71	309
N			2	2
Periphyton	1.0148	0.9091	89.6	6,326
	0.5763	0.5038	87.4	5,803
$\bar{X}$			88.5	6,065
SD			1.56	370
N			2	2
May 16-20 *Cladophora*	0.4626	0.3467	74.9	5,234
	0.6311	0.4876	77.3	5,254
$\bar{X}$			76.1	5,244
SD			1.7	14.1
N			2	2

(continued)

TABLE 31, continued.

	Dry Weight (g)	AFDW (g)	% AFDW	cal/g
May 16-20	0.3773	0.2864	75.9	5,132
Cladophora + Epiphytes	0.4589	0.3490	76.1	5,198
$\bar{X}$			76.0	5,165
SD			0.14	47
N			2	2
Periphyton	1.0781	0.9610	89.1	6,181
	1.1199	0.9997	89.2	6,421
$\bar{X}$			89.2	6,301
SD			0.07	170
N			2	2

TABLE 32. RESPIRATION, 1978 FATHEAD MINNOW EXPERIMENT

	Tissue (mg) (Dry weight)		ul O_2/g/hr[1]			cal/g/hr*	cal/fish/day
	Fish	Total	$\bar{X}$	SD	N		
Periphyton							
May 16							
Tray 1							
Jar 1	0.4869						35.734
	0.3831	0.870	650.6	5.1	2	3.058	28.116
Jar 2	0.5386						40.966
	0.3396	0.878	674.3	25.1	3	3.183	25.943
$\bar{X}$						3.12	32.69
SD						0.088	6.93
N						2	4
May 17							
Tray 2							
Jar 3	0.2821						20.433
	0.2624	0.545	642.1	288.7	4	3.018	19.006
Jar 4	0.3127						15.976
	0.2205	0.533	452.9	123.4	3	2.129	11.265
$\bar{X}$							16.67
SD							4.054
N							4
May 18							
Tray 2							
Jar 4A	0.2821						25.764
	0.2624	0.545	809.7		1	3.806	23.965
Jar 5	0.3127						9.734
	0.2205	0.533	276.0		1	1.297	6.864
$\bar{X}$						2.563	16.582
SD						1.087	9.664
N						4	4
Cladophora + Epiphytes							
May 20							
Tray 3							
Jar 22	0.1263	0.126	2240	34	2	10.528	31.912
Jar 26	0.1557	0.156	1974	418	3	9.278	34.670
$\bar{X}$						9.903	33.291
SD						0.884	1.950
N						2	2

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*1 ml O_2 = 4.7 calories.

(continued)

TABLE 32, continued.

	Tissue (mg) (dry weight)		ul O_2/g/hr			cal/*	
	Fish	Total	$\bar{X}$	SD	N	g/hr	cal/fish/day
May 22							
Tray 4							
Jar 10	0.0905						17.409
	0.1102	0.201	1705.4	74.1	3	8.015	21.198
Jar 11	0.1305	0.131	2294.9	214.6	3	10.787	51.957
$\bar{X}$						9.400	30.188
SD						1.96	18.95
N						2	3
Cladophora							
May 23							
Tray 5							
Jar 13	0.1305	0.131	1404	231	3	6.599	20.667
May 24							
Tray 6							
Jar 20	0.1012						14.326
	0.1157	0.217	1255	1.4	2	5.899	16.383
$\bar{X}$							15.355
SD							1.455
N							2

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*1 ml O_2 = 4.7 calories.

TABLE 33. FOOD CONSUMPTION, 1978 FATHEAD MINNOW EXPERIMENT

	Wet Weight gm	Dry Weight gm	Ash Free Dry Weight gm [a]	No. Fish	gm AFDW consumed/ fish	cal. consumed/ fish/day [b]
Periphyton						
Mar. 11-20						
Tray 1						
$\bar{X}$	55.09	23.36	2.570	20 (3/11-3/16) 16 (3/17-3/20)	0.142	544.5
SD	20.4	8.7	0.95		0.553	
N	7	7	7		7	
Tray 2						
$\bar{X}$	59.6	25.99	2.782	20 (3/11-3/16) 15 (3/17-3/20)	0.158	605.9
SD	18.4	7.80	0.858		0.049	
N	9	9	9		9	
Mar. 21-31						
Tray 1						
$\bar{X}$	66.0	27.99	3.078	16	0.192	736.2
SD	19.1	8.09	0.890		0.056	
N	8	8	8		8	
Tray 2						
$\bar{X}$	41.2	17.49	1.924	15	0.128	490.8
SD	12.0	5.08	0.559		0.037	
N	9	9	9		9	
Apr. 1-15						
Tray 1						
$\bar{X}$	29.7	13.13	1.444	16 (4/1-4/13) 7 (4/14-15)	0.104	398.8
SD	16.5	6.31	0.694		0.058	
N	8	8	8		8	
Tray 2						
$\bar{X}$	17.1	7.25	0.798	15 (4/1-4/13) 8 (4/14-15)	0.060	230.1
SD	8.0	3.41	0.374		0.031	
N	8	8	8		8	

(continued)

TABLE 33, continued.

	Wet Weight gm	Dry Weight gm	Ash Free Dry Weight gm	No. Fish	gm AFDW consumed/ fish	cal. consumed/ fish/day
Periphyton						
Apr. 16-30						
Tray 1						
$\bar{X}$	53.8	22.81	2.509	7	0.358	1372.8
SD	17.2	7.27	0.800		0.114	
N	11	11	11		11	
Tray 2						
$\bar{X}$	39.0	16.15	1.817	8	0.227	870.5
SD	15.8	6.70	0.737		0.092	
N	12	12	12		12	
May 1-13						
Tray 1						
$\bar{X}$	47.13	19.99	2.198	7	0.314	1204.1
SD	30.18	12.79	1.407		0.200	
N	6	6	6		6	
Tray 2						
$\bar{X}$	22.7	9.63	1.060	8	0.133	510.0
SD	10.5	4.46	0.490		0.	
N	6	6	6		6	
May 14-23						
Tray 1						
$\bar{X}$	18.2	6.44	0.847	7	0.220	843.6
SD	6.4	4.66	0.298	(5/14-	0.141	
N	6	6	6	5/16)	6	
				3		
				(5/17-23)		
Tray 2						
$\bar{X}$	14.5	6.13	0.674	8	0.174	667.2
SD	6.6	2.81	0.309	(5/14-	0.129	
N	6	6	6	5/17)	6	
				3		
				(5/20-23)		
Cladophora + Epiphytes						
Mar. 11-20						
Tray 3						
$\bar{X}$	0.60	0.063	0.044	20	0.003	13.2
SD	0.40	0.042	0.030	(3/11-	0.002	
N	9	9	9	3/15)	9	
				16		
				(3/16-20)		
Tray 4						
$\bar{X}$	0.84	0.088	0.062	20	0.003	13.2
SD	0.57	0.060	0.042	(3/11-	0.002	
N	9	9	9	3/16)	9	
				16		
				(3/17-20)		

(continued)

TABLE 33, continued.

	Wet Weight gm	Dry Weight gm	Ash Free Dry Weight gm	No. Fish	gm AFDW consumed/ fish	cal. consumed/ fish/day
Mar. 21-31						
Tray 3						
$\bar{X}$	0.60	0.063	0.044	16	0.003	13.2
SD	0.31	0.032	0.023		0.002	
N	9	9	9		9	
Tray 4						
$\bar{X}$	0.59	0.063	0.044	16	0.003	13.2
SD	0.42	0.044	0.031		0.002	
N	9	9	9		9	
Apr. 1-15						
Tray 3						
$\bar{X}$	1.67	0.175	0.123	16 (4/1-4/12) 9 (4/13-15)	0.010	44.48
SD	0.95	0.100	0.070		0.007	
N	9	9	9		9	
Tray 4						
$\bar{X}$	1.44	0.151	0.106	16 (4/1-4/12) 9 (4/13-15)	0.008	35.58
SD	0.65	0.069	0.048		0.005	
N	9	9	9		9	
Apr. 16-30						
Tray 3						
$\bar{X}$	1.11	0.131	0.092	9	0.010	44.48
SD	1.38	0.144	0.101		0.011	
N	14	14	14		14	
Tray 4						
$\bar{X}$	1.35	0.142	0.119	9	0.013	57.82
SD	1.69	0.178	0.120		0.013	
N	14	14	14		14	
May 1-13						
Tray 3						
$\bar{X}$	0.41	0.067	0.047	9	0.005	22.24
SD	0.33	0.023	0.017		0.002	
N	6	6	6		6	
Tray 4						
$\bar{X}$	0.97	0.102	0.717	9	0.008	35.58
SD	0.27	0.029	0.021		0.002	
N	6	6	6		6	

(continued)

TABLE 33, continued.

	Wet Weight gm	Dry Weight gm	Ash Free Dry Weight gm	No. Fish	gm AFDW consumed/ fish	cal. consumed/ fish/day
May 14-23						
Tray 3						
$\bar{X}$	0.62	0.065	0.046	9	0.005	22.24
SD	0.07	0.008	0.005	(5/14-	0.005	
N	6	6	6	5/19)	6	
				5		
				(5/20-23)		
Tray 4						
$\bar{X}$	0.67	0.071	0.050	9	0.007	31.12
SD	0.26	0.028	0.020	(5/14-	0.002	
N	6	6	6	5/21)	6	
				5		
				(5/22-23)		
Cladophora						
Mar. 11-20						
Tray 5						
$\bar{X}$	0.07	0.013	0.010	20	0.0005	2.0
SD	0.1	0.019	0.016	(3/11-	0.0008	
N	8	8	8	3/16)	8	
				16		
				(3/17-20)		
Tray 6						
$\bar{X}$	0.05	0.009	0.008	20	0.0004	1.60
SD	0.07	0.013	0.011	(3/11-	0.0006	
N	9	9	9	3/16)	9	
				16		
				(3/17-20)		
Mar. 21-31						
Tray 5						
$\bar{X}$	0.06	0.011	0.009	16	0.0005	2.0
SD	0.08	0.014	0.012		0.0006	
N	9	9	9		9	
Tray 6						
$\bar{X}$	0.10	0.018	0.015	16	0.0009	3.59
SD	0.11	0.021	0.017		0.0011	
N	9	9	9		9	
Apr. 1-15						
Tray 5						
$\bar{X}$	0.06	0.010	0.009	16	0.0005	2.0
SD	0.10	0.018	0.015	(4/1-	0.0007	
N	11	11	11	4/13)	11	
				9		
				(4/14-15)		

(continued)

TABLE 33, continued.

	Wet Weight gm	Dry Weight gm	Ash Free Dry Weight gm	No. Fish	gm AFDW consumed/ fish	cal. consumed/ fish/day
Tray 6						
$\bar{X}$	0.18	0.033	0.028	16	0.0020	7.98
SD	0.19	0.035	0.029	(4/1-	0.0018	
N	11	11	11	4/13)	11	
				6		
				(4/14-15)		
Cladophora						
Apr. 16-30						
Tray 5						
$\bar{X}$	0.12	0.021	0.017	9	0.0019	7.58
SD	0.17	0.031	0.026		0.0029	
N	14	14	14		14	
Tray 6						
$\bar{X}$	0.05	0.009	0.008	6	0.0013	5.19
SD	0.07	0.012	0.010		0.0017	
N	14	14	14		14	
May 1-23						
Tray 5						
$\bar{X}$	0.019	0.003	0.002	9	0.0003	1.2
SD	0.037	0.006	0.005		0.0006	
N	9	9	9		9	
Tray 6						
$\bar{X}$	0.02	0.004	0.003	6	0.005	20.0
SD	0.04	0.007	0.005		0.009	
N	8	8	8		8	

[a] % ash free dry weight, Table 34

[b] Based on Table 34
Periphyton - 3834.6 cal./gm
Cladophora + epiphytes - 4408 cal./gm
Cladophora - 3991.4 cal./gm

TABLE 34. DATA FOR BASIS OF CONVERSIONS FROM DRY WEIGHT TO CALORIES, 1978 FATHEAD MINNOW FOOD EXPERIMENTS

	Dry Weight (g)		Ash Free Dry Weight (g)		% Ash Free Dry Weight		Ash Free Dry Weight cal/g	
Periphyton	1.465	0.830	0.130	0.108	8.9	13.0	3690.9	4003.6
	1.319	1.027	0.123	0.134	9.3	13.0	4205.9	3184.2
	1.109	0.908	0.102	0.116	9.2	12.7	3486.1	4074.5
							3959.5	4072.2
$\overline{X}$					11.02		3834.6	
SD					2.07		350.7	
N					6		8	
Cladophora + Epiphytes	0.452	0.526	0.321	0.349	71.0	66.4	4480.0	4308.0
	0.408	0.566	0.307	0.375	75.3	66.3	4358.0	4470.0
	0.494	0.513	0.373	0.345	75.5	67.2	4167.0	4379.0
							4406.0	4699.0
$\overline{X}$					70.30		4408.4	
SD					4.32		153.6	
N					6		8	
Cladophora	0.147		0.385		82.1		4044.7	4036.3
	0.182		0.400		80.2		4190.9	3835.0
	0.150		0.251		86.0		3648.9	4022.1
							4115.4	4051.9
							3977.8	
$\overline{X}$					82.77		3991.4	
SD					2.96		160.7	
N					3		9	

TABLE 35. UTILIZATION OF CALORIES PER FATHEAD MINNOW

	Tissue $\bar{X}$ cal/ Fish	Gain or Loss	Resp. $\bar{X}$ cal/ Fish	Total cal. Assim.	% Wt. Gain or Loss	% Resp.	Total Food Ingest.	% Wt. Gain or Loss	% Resp.	% Ingest. Assim.
Periphyton										
Tray 1										
March 7	1700									
April 14	2020	320	1242.1	1562.1	20.5	79.5	20,760	1.5	6.0	7.5
May 23	2469	769	2517.1	3286.1	23.4	76.6	45,080	1.7	5.6	7.3
Tray 2										
March 7	1148									
April 14	1327	179	631.9	810.9	22.0	78.0	16,497	1.1	3.8	4.9
May 23	1520	372	1280.5	1652.5	22.5	77.5	26,590	1.4	4.8	6.2
Cladophora + Epiphytes										
Tray 3										
March 7	1163									
April 14	546	(-617)	1265.0	1882.0	32.8	67.2	940	65.6	134.6	200.2
May 23	288	(-875)	2563.0	3438.0	25.5	74.5	1223	71.5	209.6	281.1
Tray 4										
March 7	1694									
April 14	866	(-826)	1147.1	1973.1	41.9	58.1	815	101.4	140.8	242.1
May 23	487	(-1207)	2324.5	3531.5	34.2	65.8	1677	72.0	138.6	210.6
Cladophora										
Tray 5										
March 7	1253									
April 14	710	(-543)	785.4	1328.4	40.9	59.1	76	714.5	1033.0	1747.8
May 13	330	(-923)	1591.4	2514.4	36.7	63.3	131	704.6	1214.8	1919.4
Tray 6										
March 7	1522									
April 14	924	(-598)	583.5	1181.5	50.6	49.4	172	347.7	339.2	686.9
May 13	440	(-1078)	1182.3	2260.3	47.7	52.3	101	1067.3	1170.6	2237.9

TABLE 36. RADIOACTIVITY IN BODY PARTS AND CALCULATED ASSIMILATION RATIOS ON SUCCESSIVE DAYS FOR FATHEAD MINNOWS FED THREE DIETS

Diet	5/25						5/26					
	Fish No.	DPM/mg AFDW in: Musculature*	DPM/mg AFDW in: Viscera	DPM/mg AFDW in: Gut	Assimilation Ratio: Musculature / Gut	Assimilation Ratio: Musculature & viscera / Gut	Fish No.	DPM/mg AFDW in: Musculature*	DPM/mg AFDW in: Viscera	DPM/mg AFDW in: Gut	Assimilation Ratio: Musculature / Gut	Assimilation Ratio: Musculature & viscera / Gut
Cladophora	1	4.61	--**	5.48	0.84***	--	1	1.19	1.10	2.58	0.46***	0.96***
	2	0.03	--	1.33	0.02	--	2	88.8	--	553.7	0.16	--
	3	1.72	--	1.26	1.37	--	3	192.0	--	50.0	3.84	--
							4	169.0	9.60	80.2	2.10	2.22
Cladophora + Epiphytes	1	161.0	61.9	732.7	0.22	0.30	1	165.3	--	388.0	0.43	--
	2	0.0	--	701.4	0.0	--	2	243.5	133.3	2376.7	0.10	0.16
	3	45.7	--	249.3	0.18	--	3	213.5	91.6	1810.6	0.12	0.17
	4	51.4	15.0	299.4	0.17	0.22	4	180.2	251.2	1519.9	0.12	0.28
					0.14±0.10	0.26±0.06					0.19±0.16	0.20±0.07
Periphyton	1	384.5	--	384.5	1.00	--	1	1167.3	2096.0	1989.2	0.59	1.64
	2	87.1	--	132.8	0.66	--	2	512.3	46.8	1533.2	0.33	0.37
					0.83±0.24		3	463.9	589.3	1385.9	0.34	0.76
											0.42±0.15	0.92±0.65

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* Head with gills not included

** Missing data

*** Range on ratios too great to warrant calculation of mean.

TABLE 37. PARTIAL RADIOACTIVITY BUDGETS FOR DIET STUDIES OF FATHEAD MINNOWS

Diet	5/24 DPM Introduced	5/25 DPM Removed		% added DPM ÷ No. fish	% DPM Ingested	DPM Introduced	5/26 DPM Removed		% added DPM ÷ No. fish	% DPM Ingested
Cladophora										
	5,151,867	Feces	103	0.0003		4,849,455	Feces	3,662	0.019	
		Fish (7)	1,531	0.0042			Fish (4)	13,752	0.071	
		Muscle	[illegible]97				Muscle	3,043		
		Heads	985				Heads	1,557		
		Guts	149				Guts	9,104		
		Viscera	(0)				Viscera	(48)		
					I* 0.0049					I 0.066
					A**0.0077					A 0.016
Cladophora + Epiphytes	5,062,182	Feces	39,124	0.097		4,817,058	Feces	24,592	0.128	
		Fish (8)	31,213	0.077			Fish (4)	58,626	0.305	
		Muscle	8,406				Muscle	11,399		
		Heads	3,936				Heads	7,475		
		Guts	18,242				Guts	38,692		
		Viscera	629				Viscera	1,060		
					I 0.142					I 0.328
					A 0.022					A 0.065
Periphyton	3,973,239	Feces	11,087	0.056		3,666,873	Feces	8,696	0.080	
		Fish (5)	109,512	0.551			Fish (3)	198,323	1.800	
		Muscle	50,005				Muscle	87,696		
		Heads	18,616				Heads	33,978		
		Guts	40,891				Guts	62,686		
		Viscera	(-)				Viscera	13,963		
					I 0.262					I 0.649
					A 0.252					A 0.924

* I = Feces and guts; ** A = Muscle and Viscera

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II. CHEMICAL COMPOSITION OF CLADOPHORA GLOMERATA (L.) Kütz. CELLS

INTRODUCTION

Experiments in which many possible consumer organisms were fed *Cladophora glomerata* showed that the alga was either not eaten or caused deleterious physiological changes.

The nutritive quality of a food depends on many factors, including not only its biochemical composition but also the nutritional requirements of the consumer species. Algae of similar ultimate energy content (caloric value) may differ widely as food sources because of variations in protein, lipid, or vitamin concentration, or differences in content of nutritionally important mineral elements. In addition, textural, gustatory, and toxic factors may be important in governing feeding behavior.

The first line of defense for an organism which may be eaten is its exterior layer; in the case of algae, this means its cell wall. The cell walls of *Cladophora* spp. and those of other members of the Cladophorales consist largely of crystalline cellulose microfibrils laid down in a mat-like structure in two directions, lying at somewhat less than a right angle (1). This orientation is unusual and may be restricted to this and closely related orders of the Chlorophyceae. The sole hydrolysis product of this cellulose is glucose, unlike many other algal celluloses which afford large quantities of pentoses or other sugars upon hydrolysis. The walls of *Cladophora* contain other constituents also, including proteins (2) and polysaccharides containing sulfate esters (3).

Some of the proteins in *Cladophora* walls are structurally important, as shown by studies which showed significant weakening of wall strength after treatment with proteolytic enzymes or disulfide reducing agents (4).

It is not known whether the unusual structure of *Cladophora* walls contributes to its resistance to predation.

The question of the existence of feeding inhibitors or toxins in *C. glomerata* has not been fully studied. However, it has been reported that several green algae, including *Cladophora* species (5; LaLonde, personal communication) contain substances toxic to mosquito larvae, and a marine *Cladophora* produces acrylic acid (6), an irritating substance with antibiotic activity. Several other bloom-forming algae, particularly blue-greens (7), produce toxic compounds; *Chara* and *Nitella* appear to contain mosquito larvicides (8); and several planktonic algae are rejected by filter-feeding grazers, notably Daphnia, presumably on the basis of taste (9).

In this section of the report we summarize analyses of the chemical composition of Cladophora, with emphasis on possible nutritionally important factors.

METHODS AND PROCEDURES

Nitrogen Compounds

Proteins--Total protein was determined by a modified Lowry (10) procedure. Freeze-dried algal samples (100-500 mg) were ultrasonically homogenized with ice cooling in 10 ml of 0.02 M phosphate buffer (pH 7.0). After centrifugation (500 rpm) of the homogenate, protein was precipitated from the supernatant liquid by using an equal volume of 10% trichloroacetic acid. After further centrifugation, the supernatant was removed for soluble carbohydrate and amino acid determinations, and the pellet was redissolved in 1.0 ml of phosphate buffer. A 1.0-ml portion of the Lowry $CuSO_4$-tartrate reagent was added, followed after 10 minutes by 3.0 ml of 2N phosphomolybdate-phosphotungstate ("Phenol reagent solution," Fisher Scientific Co.). The color was allowed to develop for 45 minutes and absorbance was read at 540 nm. Bovine serum albumin was used as a standard protein.

Amino acids--Free amino acid and peptide content of algal material was determined fluorimetrically using the method of Udenfriend, et al. (11). An aliquot (0.1 ml) of the supernatant from the protein determination was mixed with 2.0 ml 0.2 M phosphate buffer (pH 8.0), 0.2 ml of an 0.05 M aqueous ninhydrin solution and 0.1 ml of an 0.01 M solution of phenylacetaldehyde in ethanol. After mixing, the tubes were covered with aluminum foil and heated for 15 minutes in a water bath at 60°C. After cooling in cold water for 15 minutes, the fluorescence intensity (excitation 390 nm, emission 490 nm) was read using a Turner model 430 spectrofluorometer. Solutions of 0-20 mg/l glycine in 10% trichloroacetic acid were used as standards.

Amino acid composition of algal proteins was determined by acid hydrolysis. In a typical experiment, 309 mg of Cladophora (dried at 60°C) was suspended in 6 ml of 50% (6 M) HCl, purged with N_2, and heated in a Pyrex tube sealed with a Teflon-lined screw cap for 20 hours at 110°C. The hydrolysis mixture was filtered (glass fiber filter) and the filtrate was passed over a 2 cm x 12.5 cm column of Bio-Rad AG50 (H+ form). The column was rinsed successively with three column volumes (40 ml) of deionized water and three column volumes of 3M NH_4OH. The ammonia eluate was freeze-dried and portions were converted to amino acid N-trifluoroacetyl-n-butyl esters using the procedure of Roach and Gehrke (12). The ester mixture was injected onto a gas chromatograph fitted with a glass (183 x 0.2 cm) 3% OV-210 column. The areas and retention times of the emerging peaks were determined electronically and compared to that of an internal standard (n-butyl stearate). As a check on the method, a protein of known amino acid composition (bovine serum albumin) was hydrolyzed and derivatized by the same procedure.

Lipids

Total lipids--Total lipid was determined gravimetrically by Soxhlet extraction of freeze-dried algae (usually 2-3 g) in a pre-extracted paper thimble. Chloroform-methanol (2:1 v:v) was used as solvent; a blank thimble containing no algae was used as a control.

Free fatty acids and hydrocarbons--Freeze-dried algae (3-6g) were extracted in a Soxhlet apparatus with pesticide-grade pentane. The pentane was evaporated and the total pentane-soluble lipid was determined gravimetrically, with correction for simultaneously extracted blank thimble. Fatty acids were removed by shaking the total extract three times with 25-ml portions of 1% Na_2CO_3 (pH 11). The combined Na_2CO_3 extract was back-extracted three times with pentane, acidified to pH 3 with 5% HCl, and re-extracted with pentane and evaporated to obtain the total free fatty acids. The weight of this fraction was determined; a portion was removed for yeast toxicity studies, and the remainder was esterified using absolute methanol containing 12% redistilled $BF_3 \cdot Et_2O$ (13). For each sample, 3.0 ml of the reagent was added and the solution was boiled for 3 minutes; after cooling 6 ml water and 3 ml pesticide-grade petroleum ether was added. The top (petroleum ether) layer was removed and concentrated to about half its volume under a stream of nitrogen. After accurate volume determination (calibrated microsyringe), the solution was transferred to a GC vial and a 4.5-ul portion of the sample was injected onto a 10% diethylene glycol succinate column (glass, 200 x 0.2 cm). The GC oven temperature was programmed from 80° to 190°C at 6°/min. The areas of peaks emerging from the column were electronically integrated. Each sample was injected three times for determination of peak areas and relative retention times. A mixture containing known amounts of C_8-C_{18} fatty acid methyl esters (Sigma Chemical Co.) was used to identify and quantitate the individual fatty acids. Pentane-soluble (non-fatty acid) material remaining after carbonate extraction was weighed, used for yeast toxicity studies, and also partly characterized by ultraviolet and infrared spectra.

Sterols--Sterols were isolated from the total lipid fraction by saponification (25% methanolic KOH: reflux 45 min.), extraction with ether, and precipitation from the nonsaponifiable fraction with digitonin. Recovery of free sterols from the digitonide precipitate was done by the method of Issidorides, et al.(14); the crude precipitate was heated with dimethylsulfoxide and extracted with hexane. The hexane extract was dried (Na_2SO_4), filtered and evaporated to dryness. An aliquot of the sterol fraction was injected into a 3% OV-210 glass GC column held isothermally at 250°C. Under these conditions, authentic cholesterol emerged at 9.5 minutes.

The total lipid fraction of Cladophora and its nonsaponifiable portion were also examined by thin layer chromatography on silica gel. The solvent used was 1/1/0.02 hexane/ether/acetic acid,and the spots were visualized by spraying with 5% H_2SO_4 in methanol followed by charring in a 120°C oven. Authentic cholesterol gave a red spot at R_f 0.45 when run under these conditions.

Soluble Carbohydrates

Soluble carbohydrate in algal samples was determined using the supernatant material from the protein determination. The phenol-sulfuric acid colorimetric method of Dubois, et al. (15) was used. Standard solutions used consisted of glucose in 5% trichloroacetic acid.

Total Phosphorus

A persulfate digestion method was used. Determinations were performed on accurately weighed freeze-dried samples of about 1 mg dry weight. Finely snipped algal fragments were suspended in 50 ml deionized water, 5 ml of 5.4 M H_2SO_4 and 7.5 ml of 0.18 M $K_2S_2O_8$ were added, and the mixture was digested in an autoclave (1.4 kg/cm^2) for 30 minutes. After cooling, the samples were brought just to the phenolphthalein end point and diluted with deionized water to exactly 100 ml. The phosphate concentration in a 50-ml subsample was determined by the ascorbic acid method (16).

Toxicity Studies

Preliminary feeding experiments with snails presented problems which made the results difficult to interpret. We therefore decided to carry out a series of experiments on yeasts.

Yeast screening experiments--In preliminary studies designed to identify the chemical nature of toxic materials *Cladophora* might contain, a yeast growth inhibition test was used. Yeast, rather than some other microorganism, was chosen because of its eukaryotic nature; it is similar in its cellular organization to higher animals, and most materials toxic to yeast are also toxic to higher forms. The method has been briefly outlined (17). Freeze-dried yeast (90% viable *Saccharomyces cerevisiae* Sigma) was grown in a defined basal medium containing 1% glucose as sole carbon source (18) to the midexponential growth phase. A portion of this suspension containing (1+ 0.1) x 10^6 cells was removed and added to fresh medium containing the test material. Six replicate cultures were incubated at 25°C using a reciprocating water-bath shaker for 48 hours. Growth was measured turbidimetrically and expressed as a percentage relative to six control cultures. Differences in growth were assessed for significance using Student's t-test.

RESULTS

Nutrient Content

Proteins--It has been suggested that the amounts of protein and component amino acids are the primary determinants of the food quality of plant materials (19). The protein content of algae is extremely variable, with levels from 5% to 60% being reported in different species. Within an individual species there can also be great variations in protein composition depending on light, temperature, phase of growth, and other factors. Hare and Schmidt (20) showed a fourfold variance in protein content in synchronized

Chlorella cultures, with young and rapidly growing cells having the highest levels of protein.

Our results on *C. glomerata* indicate that this alga contains only about 8% protein (Table 1). It is likely that this is partly due to its filamentous growth habit and to the presence of a thick, largely cellulosic cell wall. The data of others, as well as our own determinations on *Vaucheria* and *Stigeoclonium* spp. show that filamentous algal forms are low in protein relative to other types. Literature data for diatoms, and our analysis of a diatom-dominated mixed periphyton community readily acceptable to a variety of consumer organisms, indicate that a much higher level (over 40% protein) is characteristic of these algae.

The possibility was considered that *Cladophora* protein might also be deficient in some essential amino acid constituents. Although the amino acid requirements of most aquatic invertebrates are not very well understook, most insects are known to require branched-chain (isoleucine, leucine, valine), basic (lysine, arginine, histidine), and aromatic (phenylalanine, tyrosine, tryptophan) amino acids. *Cladophora* was hydrolyzed in strong acid and its amino acid constituents were concentrated and analyzed by GC. As a check on the applicability of the method, a standard protein (bovine serum albumin) whose amino acid composition has been determined by traditional ion-exchange chromatographic procedures was hydrolyzed, derivatized, and analyzed. The individual amino acid percentages computed using the GC method were compared with those from the literature by linear regression; a high degree of correlation ($r=0.940$, $P \leq 0.001$) was demonstrated.

The amino acid composition of *Cladophora* protein and that of some other algae is summarized in Table 2. As the table indicates, the amino acid composition of *Cladophora* protein was not exceptional and would appear to be adequate to provide for the nutritional requirements of invertebrates. Aromatic, basic, and branched amino acids were all represented. Tryptophan was destroyed by the hydrolytic method used and arginine and histidine were not detected by GC; however, these amino acids make up only about 5% of a typical protein. Some insects require methionine, and although *Cladophora* protein appeared to be low in this amino acid, other freshwater algae contained even less of it. The apparent absence of cysteine, another sulfur-containing amino acid, is interesting; but this amino acid was low in the proteins of some other freshwater algae as well. *Cladophora glomerata* appears to differ considerably in this respect from *C. rupestris*, a marine form which was shown to have an exceptionally high content of sulfur amino acids (22).

The overall amino acid composition of *Cladophora* protein showed significant correlations with that of other algae; linear regression of *Cladophora* composition vs. diatom composition, for example, gave $r = 0.86$.

Free amino acids and peptides were shown to be present in a *Cladophora* extract, but the amount was low (0.37% of dry weight). This is a rather typical concentration for most biological materials, although in some cases (for example in large seeds) high concentrations of non-protein amino acids may be found; they appear to have deterrent effects on some predators (23).

Lipids--The lipids of algae make up a heterogeneous group of substances, having as common characteristics the ability to dissolve in nonpolar solvents and a high ratio of carbon to hydrogen. Their reduced character makes them a useful source of metabolic energy. Some lipids are also essential nutrients for consumer organisms; thus some invertebrates require a dietary source of polyunsaturated fatty acids, such as linoleic or linolenic acid. In addition, all insects require dietary sterols. The requirement for sterols by other invertebrates is not as well established but it may be rather general. Many blue-green algae either contain no sterol or very low concentrations of sterols; this may explain, to some extent, why they are such poor dietary constituents for invertebrates. For green algae, the reported range of sterol content is 0.01 - 0.38% (25, 26). The only member of the genus Cladophora previous studied, C. flexuosa, contained a rather low 0.06% sterol, with the major constituents being cholesterol, 24-methylene-cholesterol, and 28-isofucosterol (26). Other members of the Cladophorales also have been reported to produce sterols; for example, Chaetomorpha crassa contains sitosterol, cholesterol, 24-methylenecholesterol, and two methylated sterols (27).

The combined fatty acids of algal lipids have been extensively studied. Although there is considerable variation among individual genera, larger groups of algae are characterized by more or less typical fatty acid patterns. For example, the usual fatty acid composition of diatoms is dominated by unsaturated C_{16} and C_{20} fatty acids. Unsaturated C_{18} fatty acids, particularly linolenic acid, are normally present at much lower levels (21). Green algae, on the other hand, resemble the higher plants in their fatty acid composition; their unsaturated fatty acids are usually C_{18} types, with unsaturated C_{16} or C_{20} acids present in low concentration (28).

Much less work has been reported on the free fatty acids of algae, although there are a few interesting observations on their toxicity to other algae (29), protozoa (30), and bacteria (31).

There are a considerable number of other reports of lipid toxicity. Polar lipids, in particular, may interact with cell or organelle membranes and cause disorganization of systems involved in transport, photosynthesis, or respiration. Surfactants, such as linear alkylsulfonates, have similar toxic effects on aquatic organisms for related reasons. Restricting consideration only to simple fatty acids and their esters, there are reports of toxicity to bacteria (32), fungi (33), and algae (34) in addition to those mentioned previously. Although toxicity of C_{16} - C_{20} fatty acids and unsaturated fatty acid peroxides (35, 36) has been mentioned by a few investigators, there are many more reports of activity in the C_8 - C_{14} range of saturated acids with a peak at about C_{12}.

The total lipid content of Cladophora (about 9.2%; Table 1) was somewhat less than that of mixed periphyton (10.4%), other diatoms (about 13%), or Vaucheria (12.2%). Examination of the total lipid extract by thin-layer chromatography revealed two closely spaced bands, running at practically the same R_f as an authentic sample of cholesterol. The lipid fraction was fractionated by means of digitonin to obtain the sterols; decomposition of the digitonide complex gave a crude sterol fraction amounting to 0.13% of the total weight of the Cladophora. This sterol content is in the range reported for other green algae, and is about twice the amount found in Cladophora flexuosa (25). The crude sterol fraction, when examined by GC, gave only a single broad peak, eluting at the same time as cholesterol, which was previously reported as a constituent of algae of this order. Further study will be required to confirm this tentative identification.

The free fatty acid content of Cladophora was investigated because of the several reports concerning potentially toxic free fatty acids in other algae. Similar studies were performed on a mixed periphyton community (dominated by diatoms) which was readily eaten by the snail Physa. The algae were extracted with refluxing pentane and the total extract was partitioned into carbonate soluble (free fatty acids) and insoluble (hydrocarbon, etc.) portions. The free fatty acid fraction was examined by GC and the results are shown in Table 3. Periphyton contained more free fatty acids, with the majority consisting of C_{14} and C_{16} isomers. Cladophora contained mostly C_{18} - unsaturated fatty acids. These data are consistent with other reports of the relative abundances of fatty acids in the triglycerides of diatoms and green algae, respectively.

Short-chain free fatty acids were also found in these algae. Both contained some C_8, but Cladophora contained 170 ppm C_{12} (lauric) acid, which was absent in the periphyton. Conversely, C_{10} (capric) acid occurred in periphyton but not in Cladophora. LaLonde (personal communication) has independently shown that Cladophora contains lauric but not capric acid. The occurrence of lauric acid in Cladophora is interesting from an ecological standpoint since there are many reports of its toxicity; these will be discussed under toxicity experiments.

The pentane-soluble, carbonate-insoluble fraction of Cladophora was examined by infrared and ultraviolet spectra; it included hydrocarbons and long-chain alcohols as well as some carbonyl-containing material.

Carbohydrates--The bulk of the carbohydrate in most algal cells is polymeric, with a majority in many forms (green, brown, red, and some yellow-green algae) being cellulose. Because cellulose is not digestible by most animals, requiring a specialized gut microflora such as that found in ruminant animals, it may contribute little or nothing to the nutritional value of a given alga although it may contribute a significant number of calories as measured by combustion. Since Cladophora is known to have a cellulosic cell wall (1), we chose to measure only soluble carbohydrates, assuming these to be more available to digestion by a wide range of predators. Although little quantitative data on the soluble carbohydrate concentrations of algae have been reported, a great variety of mono- and oligosaccharides have been reported to occur, along with starches, agar-like gels, and sugar sulfonates. Several species of the Cladophorales, including Cladophora spp., produce unusual fructose-containing oligosaccharides (37). When used as storage products, as in the brown algae, soluble carbohydrate may comprise as much as 50% of algal dry weight (38).

C. glomerata contained about 4.5% soluble carbohydrate (Table 1), more than was found in the filamentous xanthophyte Vaucheria, but less than was present in a mixed periphyton (diatom-dominated) community. Because both Vaucheria and periphyton were readily eaten, it is unlikely that Cladophora was not consumed because of an insufficiency of carbohydrate. Further studies of Cladophora carbohydrates may be necessary to establish their roles in nutrition and feeding behavior. Although toxic carbohydrates are unusual, they do vary widely in palatability and susceptibility to enzymatic degradation. Starches are also feeding stimulants for a slug (39).

Total phosphorus--the phosphorus content of Cladophora (Table 1) is about 0.75%, a value intermediate between that for Spirogyra (0.19%) and periphyton (1.85%). The extent of availability of the phosphorus as a nutrient mineral for consumer organisms was not assessed.

Toxicity Experiments

Cladophora extracts (and some corresponding extracts of the readily eaten periphyton) were screened for possible toxic substances. The effect of the extract on growth of a yeast (Saccharomyces cerevisiae) in a defined medium was used for the analysis (17). Solvents used for the extraction were pentane, acetone, methanol, and cold and hot water. Results of the studies are shown in Table 4.

Water and acetone extracts of Cladophora significantly promoted the growth of Saccharomyces, probably due to the provision of respirable substrates, co-enzymes or vitamins. Methanol extracts had a limited growth-inhibiting effect; at 800 ppm growth of the treated cultures was significantly less than that of controls. Pentane extracts of Cladophora had little effect at 100 or 200 ppm, but significantly suppressed growth at 400 ppm. Pentane extracts of periphyton promoted growth significantly.

In a preliminary experiment, it was determined that the observed toxicity of the Cladophora extract was not due to its pentane-soluble hydrocarbon constituents. Yeasts exposed to this fraction (200-400 ppm) grew at 97-105% of the control growth. Because insufficient material was available, it was not possible to test the free fatty acid fraction of Cladophora for toxicity. However, LaLonde (personal communication) has shown using pure compounds that some of the free fatty acids of Cladophora are toxic to mosquito larvae, with the C_{12} (lauric), C_{14} (myristic) and $C_{16.1}$ (palmitoleic) acids being most active. Our work shows that the latter two acids are more abundant in the readily eaten mixed periphyton than in Cladophora (Table 3), and that lauric acid, absent from periphyton, is present in Cladophora. Our tentative conclusion is that lauric acid is responsible for the toxicity of Cladophora extracts to yeast, and may also contribute to Physa toxicity.

Further study will be required to confirm this hypothesis. It is interesting that lauric acid, extracted from the seaweed Ulva lactuca, was shown to be toxic to protozoa, causing cell lysis at 250 ppm and inhibiting mobility at lower concentrations (less than 100 ppm; 30). There are also many reports of antimicrobial activity of lauric acid (40, 34, 41, 42, 32, 43).

The results of these experiments show that Cladophora glomerata is lower in overall nutritive value (less protein, lipid, and phosphorus per unit of dry weight) than many other readily eaten algae. Cladophora contains low concentrations of steroids and free polyunsaturated fatty acids. Although Cladophora is also low in sulfur-containing amino acids, it is not unique in this respect, and appears to contain most of the other essential amino acids in adequate quantities. Cladophora has a thick and resistant cell wall, which affords mechanical protection and may also be resistant to enzymatic attack.

The evidence suggests that Cladophora glomerata may contain substances which are toxic based on yeast growth inhibition. One of these substances may be free lauric acid, which is present in the organism at a concentration of about 170 ppm.

TABLE 1

NUTRIENT CONTENT OF FILAMENTOUS ALGAE, PERIPHYTON, AND DIATOMS[a]

	Cladophora glomerata	*Vaucheria*	*Stigeoclonium*	*Spirogyra*	Mixed periphyton	Diatoms[b]
Protein	8.2±1.6(9)	5.4(2)	9.67(1)	15.93[c]	44.3(1)	46.4±10.4(8)
Free amino acid	0.37(2)				0.65(1)	
Sol. carbohydrate[d]	4.5±1.6(5)	1.6(1)			8.1(2)	21.6±9.1(8)[e]
Total lipid	9.2±3.0(5)	12.2(1)			10.4(1)	13.0±8.4(8)
Pentane-soluble	0.86(2)				1.11(1)	
Free fatty acid	0.15(2)				0.96(1)	
Sterol	0.13(2)					
Total P	0.74±0.35(4)	1.90(2)	1.12(2)	0.19(2)	1.85(2)	

[a] All values in percent of ash-free dry weight. Figures in parentheses are number of determinations. Standard deviations are given where this figure is greater than 3.

[b] Recalculated from data in Table 7.1 of Darley (21).

[c] Unidentified species (19).

[d] Soluble in cold 5% trichloroacetic acid.

[e] Total carbohydrate.

TABLE 2

AMINO ACID COMPOSITION OF PROTEINS OF SOME FILAMENTOUS ALGAE AND DIATOMS

Amino Acid	Cladophora[a]	Stigeoclonium[b]	Vaucheria[b]	Spirogyra[c]	Diatoms[d]
Alanine	11.02±0.82	14.9	13.1	15.04	8.2
Glycine	9.77±0.63	17.6	12.6	7.35	10.5
Valine	8.99±1.77	6.2	6.6	4.71	6.2
Leu-ileu-thr[e]	20.04±0.67	18.1	20.8	14.79	20.1
Serine	4.98±1.40	7.2	6.3	4.49	9.0
Cysteine	(0.00)	0.5	1.0	0.22	0.5
Proline	7.72±0.35	7.6	7.1	3.97	6.5
Methionine	2.39±0.71	1.1	3.0	1.50	1.8
Phenylalanine	6.92±1.51	4.6	4.3	5.28	4.4
Aspartic Acid	8.58±0.96	6.6	7.5	8.69	12.0
Glutamic Acid	7.90±0.60	6.9	8.1	18.56	12.4
Tyrosine	3.22±0.32	2.7	2.9	3.41	2.7
Lysine	8.44±0.77	6.2	6.7	5.78	4.0

[a] Mean and standard deviation of four determinations.

[b] Mean of two determinations.

[c] Boyd, 1973.

[d] Mean for cell contents of six species (24)

[e] Not always resolved by GC method used.

TABLE 3

FREE FATTY ACIDS OF CLADOPHORA AND MIXED PERIPHYTON

Acid	Cladophora	Periphyton
C_8 (caprylic)	30[a]	420
C_{10} (capric)	0[b]	170
C_{12} (lauric)	170	0[b]
C_{14} (myristic)	180	1560
C_{16} (palmitic)	70	3020
$C_{16.1}$ (palmitoleic)	40	1370
C_{18} (stearic)	75	200
$C_{18.1}$ (oleic)	105	470
$C_{18.2}$ (linoleic)	200	1280
$C_{18.3}$ (linolenic)	520	230

[a] All values in micrograms per gram ash-free dry weight (ppm).

[b] None detectable by GC.

TABLE 4

GROWTH OF SACCHAROMYCES CEREVISIAE EXPOSED TO ALGAL EXTRACTS

Alga[a]	Solvent	Concn. (ppm)	Growth[b]	Sig.[c]
C	Pentane	100	98	-
		200	115	-
		400	3	+
P	Pentane	100	152	+
		200	154	+
		400	157	+
C	Acetone	200	203	+
		400	151	+
C	Methanol	200	86	-
		400	85	-
		800	53	+
C	Water (hot)	200	200	+
		800	202	+
C	Water (cold)	200	161	+
		400	165	+
		800	183	+

[a] C = *Cladophora glomerata* : P = mixed periphyton.

[b] Growth relative to a control (only solvent added). Control growth = 100. No growth = 0. Usually mean of 6 replicates, except for pentane extracts where limited quantity of material allowed only duplicates.

[c] + = Significant at $P \pm 0.05$ (t-Test) relative to control.

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III. A FUNGAL PATHOGEN FOR POSSIBLE BIOLOGICAL CONTROL OF CLADOPHORA GLOMERATA

INTRODUCTION

These experiments were performed to explore the potential of fungal pathogens for biological control of Cladophora glomerata. Considerable research has been conducted concerning the biological control of aquatic macrophytes, such as Alternanthera philoxeroides (alligator weed) and Eichornia crassipes (water hyacinth), using both insect and microbial pathogens. In contrast, the potential for biological control of algal growths has been only superficially explored. Because Cladophora growths are problematical in the Great Lakes, other lakes and ponds, streams and brackish water environments we initiated a search for a microbial pathogen of the alga. We began with the fungi because of the demonstrated utility of some for the control of macrophytes and because several fungi were reported associated with Cladophora in the early literature.

LITERATURE REVIEW

Historically, control of algal growths has been accomplished using copper sulfate (1), but effects on other organisms and copper accumulation in sediments are undesirable complications. Herbicides have been used in some instances (2, 3, 4) but in addition to lack of specificity, resistant algal strains may develop, and the herbicides may be expensive. Control of nutrient inputs will reduce algal biomass (5, 6). However, depending on the degree of reduction and prior storage in the system, biomass accumulation may still occur, albeit to a lesser extent, if the dominant algae are species not readily consumed by herbivores. Manipulation of micronutrient metal concentrations has been suggested as a possible means of altering algal community structure to favor species desired by herbivores, resulting in more efficient transfer of energy in the food web (7 , 8). This innovative idea needs further testing before its utility can be assessed.

Biological control measures also offer the potential of control of selected algal species or groups. A number of observations have been made of viruses, bacteria, and fungi that have antagonistic activity against algae of many types (Table 1). Viral and bacterial agents have been most often associated with blue-green algae (Cyanobacteria). Most fungal-algal associations have involved Phycomycete fungi, particularly the chytrid group. However, biological control programs using these agents have yet to be developed.

Some associations between fungi and *Cladophora* species reported in the early literature were saprophytic (*Diplophylyctis laevis*, 9); *Chytridium aggregatum*, 10), but some were parasitic (*Phlyctidium spinulosum* and *Phlyctochytrium zygnematis*, 11; *Myzocytium proliferum*, and *Achlyogeton entophytum*, 12), or unclassified (*Phlytochytrium quadricorne*, 11). Canter (13) and Sparrow (14) later identified the *Myzocytium* species noted by Martin on *Cladophora* as *M. megastomum* and Dogma (15) recently found *M. megastomum* on *Cladophora* from a stream in the Philippines.

It is noteworthy that some fungi show considerable promise in programs to control the development of some aquatic macrophytes. *Acremonium zonatum* (16), *Cercospora rodmanii* (17), and *Rhizoctonia* sp. (18, 19) in particular have demonstrated utility for the control of water hyacinth (*Eichornia crassipes*) in the greenhouse and in some instances in field tests (20). Many toxins produced by fungi are host-specific (21), which makes them particularly promising biological control agents.

METHODS AND PROCEDURES

Isolation, Culture, and Identification of Fungi

Cladophora was collected from three streams where large populations were present for several months of the year (Plum Run, Chester County, Pennsylvania; Lititz Creek and Cocalico Creek, Lancaster County, Pennsylvania). Collections were examined microscopically for the presence of fungi associated with the *Cladophora*. A fungal-infested *Cladophora* stock culture was also sampled for fungal isolates.

Small pieces of *Cladophora* with associated fungi were transferred to plates of autoclaved *Cladophora* agar consisting of 6 g *C. glomerata* in 190 ml water (macerated in a blender) and 1.5 g Prepurified agar (Difco, Detroit, Michigan). Cultures were incubated at room temperature for 7-21 days to permit fungal development. Isolates were purified using malt extract broth and agar as well as chitin and cellulose (both identified in early studies of the *Cladophora* cell wall (22), although the presence of chitin has not been substantiated) in mineral salts solution (23). Stock cultures were maintained on malt extract agar and on *Cladophora* agar at 10°C.

Identification of one isolate as *Acremonium kiliense* Grutz (Fungi Imperfecti) was achieved using slide culture techniques and microscopic observations. The keys of Barnett (24), von Arx (25), and Gams (26) were used. We gratefully acknowledge Dr. Walter Gams, Institut fur Schimmelspilze, Baarn, the Netherlands, who confirmed our generic identification and affixed the species epithet for us.

Throughout all phases of this work any material exposed to any fungal agent was sterilized by autoclaving or by the addition of formaldehyde (where large volumes might preclude autoclaving) before discarding.

Culture of C. glomerata

Unialgal C. glomerata was grown in the defined Cladophora Medium II of Fitzgerald and Gerloff (27) at 20°C using a 16-hour photoperiod with average illumination intensity of 55 u Einsteins/m^2/sec from Vita-Lites (Duro-Test Corp., North Bergen, New Jersey). These lamps possess a spectrum approximating solar radiation. Cultures were bubbled continuously with filtered air.

Laboratory Assay of Fungi for Anti-Cladophora Activity

Fungi were grown on malt extract, chitin, or Cladophora substrates, usually for 7-15 days at 18-20°C. Conidia were collected from solid substrates by rinsing with 5 ml sterile water and centrifuging the suspension. Conidia were retrieved from liquid cultures containing chitin or Cladophora by centrifugation of the liquid portion of the medium; the mycelium remained largely with the solid added substrate. Numbers for addition were estimated by direct microscopic counts using a Petroff-Hausser cell. Viable numbers reported here were determined from serial dilutions spread on plates of malt extract agar in triplicate. Growth foci were enumerated after 5 and 7 days' incubation at 20°C.

The C. glomerata used in assays was either subcultured in Cladophora Medium II for 5-7 days or was collected from a field site just prior to use. Material from the field was dominated heavily by Cladophora, but diatoms, other green, and blue-green algae were present in small amounts.

Fungal-algal interaction was studied in 125 ml Erlenmeyer flasks by adding serial dilutions of A. kiliense conidia suspended in water to 25 mg (wet weight) C. glomerata in 50 ml filter sterilized (0.45 um pore size) Lititz Creek water. Controls received no addition. Cultures were incubated for 10-14 days with continuous agitation under Vita-Lites (72 u Einsteins/m^2/sec intensity at the flask surface; 16-hour photoperiod) at 20°C.

Three measures were used to assess fungal effects on Cladophora: cell condition, chlorophyll a content, and dry weight. The algae were examined microscopically for cell condition. At least 1000 cells from each flask counted and scored as healthy (packed with green chloroplasts), partially bleached (chloroplasts a pale green or fewer in number than in the control), chlorotic (chloroplasts a pale yellow-grey) or empty (cell contents absent). Counts were made across each microscopic field, rather than along Cladophora filaments. The algae from each flask were blotted dry, weighed, macerated, and extracted overnight in 10 ml acetone at 4°C for determination of chlorophyll a (28). The algae were then dried at 60°C and weighed.

Supernatants from fungal cultures or assay flasks in which effects on Cladophora were obtained were also tested for effects on Cladophora. The supernatant fluids were filter-sterilized (0.45 um pore size) and aliquots (5 ml) were added to the 45-ml filter-sterilized Lititz Creek water. Other 5-ml aliquots were heated to 80°C or 100°C for 10 minutes before addition. Cladophora (25 mg) was added, and assays were conducted as described.

Microcosm Experiments with A. kiliense

Experiments with *Cladophora*-dominated algal communities were conducted in flowing water (stream) and static (pond) microcosms in a greenhouse laboratory. Temperature was monitored using maximum-minimum thermometers (Brooklyn Thermometer Co., Farmingdale, New York). Radiation was monitored at a site outside the greenhouse using a pyranometer (Eppley Laboratories, Newport, Rhode Island). Water chemical parameters (NO_3-N, PO_4-P, and total alkalinity) were measured using Standard Methods (29), HN_3-N by the phenol-hypochlorite method (30), and metals by atomic absorption spectrophotometry.

Flowing water microcosm experiments were conducted by placing rocks with attached algal communities from Lititz Creek in Plexiglas cylinders (10 cm inside diameter x 61 cm long, fitted with end caps). The microcosms contained 2 liters Lititz Creek water and were connected to reservoirs containing 10 liters Lititz Creek water. The water in the systems was recirculated continuously using submersible pumps. The microcosms were submerged partially and the reservoirs completely in water from White Clay Creek, which flows near the laboratory. This water was continuously replenished to keep the systems at near ambient water temperatures. Every three days the water in all microcosms was renewed with Lititz Creek water and the experimental systems were reinoculated with fungal conidia (collected by centrifugation from 1-liter cultures grown on *Cladophora* in mineral salts solution) or conidia and mycelium (collected by centrifugation from cultures in malt extract broth). Cladophora condition was determined at intervals from microscopic counts of 1000 cells done on a sample taken from each rock. Additional counts of another 1000-1500 cells, which included both *Cladophora* and *Ulothrix*, were done to assess dominance and condition.

Static microcosm experiments were conducted by adding 3 grams wet weight of *Cladophora*-dominated algal communities from Lititz Creek to 6 liters of Lititz Creek water in plastic trays. The trays were kept at near-ambient stream temperatures as were the flowing water systems. The water in the trays was changed and the experimental system was reinoculated every three days. Counts of 1000 cells each were performed on duplicate samples at intervals to assess *Cladophora* condition. Another count of 1000 *Ulothrix* and *Cladophora* cells was done to assess relative dominance and condition.

RESULTS

Laboratory Assays with Isolates P-1, PR-5, and Acremonium kiliense

Fungi were observed in some *Cladophora* cells in material collected from Plum Run and isolates, designated PR-1 and PR-5, were obtained. Isolate PR-5 possessed no virulence against *Cladophora*.

In our first experiment with isolate PR-1 anti-bacterial antibiotics (Streptomycin sulfate and Penicillin G, Sigma Chemical, St. Louis, Missouri) were added to suppress bacterial numbers since axenic cultures of *C. glomerata* were unavailable. A tuft of PR-1 mycelium was added (rather than spores) to cultures in Cladophora Medium II, White Clay Creek water, or a 25% dilution of medium in water, to examine effects when nutrient concentrations were altered.

Isolate PR-1 had a slight detrimental effect on *Cladophora*, but pronounced development of another fungus occurred in the control with antibiotics (Table 2). This fungus, which was apparently associated with the *Cladophora* stock culture, was isolated and subsequently identified as *Acremonium kiliense* Grutz. Two other experiments were conducted with isolate PR-1, but it appeared less antagonistic to *Cladophora* than *A. kiliense*. *A. kiliense* (*Acremonium* was formerly known as *Cephalosporium*) therefore became the focus of further experimentation.

The results of one experiment with *A. kiliense* are shown in Figure 1. Exposure to *A. kiliense* caused chlorosis of many *Cladophora* cells. *Cladophora* collected from Lititz Creek was more susceptible to the fungus than *Cladophora* from the stock culture, perhaps because the fungus was isolated from the stock culture or because the Lititz material was a mixed algal community with other interactions. Assays were repeated several times with variable results (Table 3). Direct microscopic examination for cell condition tended to be the most sensitive measure of effect, although in experiments 5 and 6 chlorophyll *a* concentration showed the effects of fungal pathogenicity. In experiments 1-4 the number of healthy cells was reduced to less than 50% of the number in the control. (In experiments 1 and 2 50% or more of the cells at some treatment level were completely chlorotic, but in the other experiments the effect was less dramatic and the chloroplasts were partially bleached.) Lack of correlation between cell condition and chlorophyll *a* concentration when the Lititz Creek community was used may have resulted from the presence of diatoms, *Spirogyra*, *Ulothrix*, or other algae that were unaffected by *A. kiliense* and which contributed chlorophyll *a*. Sometimes microscopic observation showed ruptured cells with external accumulation of intact chloroplasts and other debris along the *Cladophora* filaments. In such instances, some chlorophyll *a* might still be measured even though the cells were debilitated.

Although the effect produced by *A. kiliense* was observed repeatedly, we have had experiments in which effects on *Cladophora* were not evidenced under the experimental conditions. Inoculum size, fungal culture conditions and age of the fungal inoculum, extent of germination of conidia, and incubation water varied between experiments, but positive or negative results cannot be traced to a single factor. Since negative results were more common in later work it is possible that loss of *A. kiliense* virulence may also be involved. Extent of conidial germination may also have affected our results so that similar inoculum levels did not produce the same result.

At the end of the experiment shown in Figure 1, supernatants from the assay flasks in which *Cladophora* was damaged (those receiving undiluted condial inocula) were assayed for effects on *Cladophora*. Only one supernatant (Lititz *Cladophora*, assay flask 1) affected the stock culture *Cladophora* strain in this experiment (Figure 2). Heating at 80°C did not eliminate the toxic effect, although exposure to 100°C may have lowered it somewhat. Variability in results may be due to differing concentrations of the toxic substance(s) in the supernatant fluids added. Furthermore, we tested only 5 ml volumes so that nutrient concentrations would not be severely altered by supernatant addition, and positive results may have been obtained with larger volumes.

In another experiment *A. kiliense* was grown in Czapek-Dox broth for five days after which the conidia and fungal culture supernatant were assayed for effects. No effect was noted from addition of conidia in this trial, but the addition of sterilized supernatant damaged *Cladophora* (Figure 3), whether the supernatant was heated or not. The control (5 ml Czapek-Dox broth) had no effect on the *Cladophora*. Another identical experiment was conducted except that the fungi were cultured for 9 days prior to addition. Only mild effects were noted, but the concentration of toxicant probably differed between experiments. Production and destruction of toxicants may be proceeding simultaneously in the fungal cultures.

These experimental results demonstrated, however, that contact between the alga and fungus was not necessary for damage but that this *A. kiliense* isolate produced an antagonistic agent against *Cladophora* that was water soluble and not destroyed by heating. This indicates that the deleterious effect of the fungus on *Cladophora* was caused by an antagonistic effect and not to competition for nutrients.

Microcosm experiments with *A. kiliense*.

Experiments were conducted with *A. kiliense* and *Cladophora*-dominated communities in microcosms held under natural light and water temperature conditions in a greenhouse laboratory. Both flowing water microcosms (designed to simulate stream conditions) and static microcosms (designed to simulate pond or lake environments) were used.

The first experiment was conducted from July 17-August 1, 1978. Light input ranged from 134-586 langleys/day ($\bar{x} \pm$ s.d. = 438 ± 126, n = 15) and temperature averaged 21.2 ± 0.98 °C ($\bar{x} \pm$ s.d., n = 15). Duplicate static systems were inoculated with 2.07 - 18.5 x 10^4 conidia/ml but effects on *Cladophora* were not evident. However, in the stream systems which were inoculated identically, the *Cladophora* was in poor condition after one week. White patches of *Cladophora* were visible to the naked eye on the surface of rocks. Direct microscopic examination indicated many cells were chlorotic in the treated microcosms, but in the controls >70% of the *Cladophora* cells were healthy (Table 4). Diatoms and *Ulothrix* which were present in the microcosms were unaffected by *A. kiliense* and protozoa and other meiofauna were healthy. Extensive *Ulothrix* development occurred in both the inoculated and control microcosms, particularly at the water surface where skeins of *Cladophora* provided sites for attachment and growth. At two weeks, counts of 1000 cells each were made of attached and surface growths. These counts included both *Cladophora* and *Ulothrix* cells. *Cladophora* cells were most often encountered in the growth scraped from rocks in all microcosms and *Ulothrix* at the water surface. Cladophora exposed to A. kiliense had many more chlorotic cells (60 and 70% of attached cells) than Cladophora in the controls (8 and 23%). Ulothrix cells were in good condition in both sets of microcosms.

The experiment was repeated between August 11-31, 1978, with striking results. Conidia added during the first week were collected from cultures grown on Cladophora in mineral salts solution used for chitin-salts medium (23). Both mycelium and conidia that were harvested from malt extract broth were added after that.

After one week, Cladophora in the inoculated streams was largely chlorotic (Figure 4), and Ulothrix was replacing Cladophora as the dominant alga (Table 5). Diatoms, Scenedesmus, and Spirogyra that were present to a smaller extent in the communities were in good condition as were protozoans and rotifers. At the end of the experiment, Cladophora was absent on some of the rocks in the inoculated microcosms. Although the condition of Cladophora in the control microcosms deteriorated through the experiment, on any date many more healthy cells were encountered in the controls than in the treated microcosms.

Treatment with A. kiliense produced similar results in the static microcosms. In one week the fungal-exposed Cladophora was largely chlorotic (Figure 5). In the controls, however, Cladophora remained healthy throughout the experiment. As in the flowing water systems, Ulothrix replaced Cladophora as the dominant alga in the treated microcosms (Table 5).

Water temperature (measured in the early morning and late afternoon) ranged between 19.5-23.0°C in the microcosms. Light inputs ranged from 169 to 584 langleys/day ($\bar{x} \pm$ s.d. = 394 ± 126, N = 21). Concentrations of selected chemical parameters in Lititz Creek water are shown in Table 6 and their concentrations in water removed from the microcosms are shown in Table 7. The lower concentrations of NO_3-N in the water removed from the treated microcosms may reflect the rapid growth of Ulothrix and diatoms in these microcosms, but PO_4-P and total alkalinity concentrations do not show the same pattern.

Negative results were obtained in trials performed in the flowing water systems between September 18-October 4 and in the static systems between September 18-October 20. Lower water temperatures (13.0 - 20.0°C in the stream systems and 15.5-20.0°C in the lake microcosms) may have prevented sufficient fungal development between changes of water and reinoculation.

A final trial was conducted between October 10-20 in the flowing water microcosms. Temperature during the experiment ranged from 9-17°C ($\bar{x} \pm$ s.d. = 13.6 ± 1.67, n=22) and light inputs from 61 to 353 langleys/day ($\bar{x} \pm$ s.d. = 252 ± 101, n=11). Water chemistry data is shown in Table 8. Conidia and mycelium were added at three inoculum levels (x, 2x, and 10x) to increase inocula over those which produced no effect in the preceeding experiments. In this experiment results were not as dramatic as during the summer experiment, perhaps because of the lower water temperature, but factors such as fungal virulence may be involved. Nevertheless, within 10 days the Cladophora in the treated microcosms was in poorer condition than that in the control microcosms (Figure 6). Ulothrix again developed larger populations in the treated microcosms, but Cladophora remained dominant in the control (Table 9).

The results presented here suggest that microbial pathogens may provide a useful tool for controlling extensive growths of C. glomerata in nature. The results of our laboratory experiments, although striking in some instances, have been variable. Nevertheless Cladophora from both stock cultures and field sites were adversely affected in laboratory experiments. Redhead and Wright (31) noted that effects of fungi on blue-green algae were least pronounced in agitated cultures when compared to effects in static cultures or in assays using discs impregnated with culture supernatant placed on algal lawns. Since all our laboratory experiments were conducted with agitation, effects have been observed under conditions considered by others as least favorable for detecting antagonism.

Effects were less pronounced in later experiments which may mean some loss of fungal virulence or increased resistence of our Cladophora stock. Redhead and Wright (31) reported A. kiliense was one of several fungi and bacteria possessing lytic activity against blue-green algae but noted their fungal isolates lost virulence after six transfers on laboratory media.

The identity of the toxic agent is unknown as yet. It is apparently non-proteinaceous because heating (which would denature proteins) did not eliminate effects. Some A. kiliense (formerly Cephalosporium acremonium) strains produce the antibiotic cephalosporin. An antibiotic-producing strain (ATCC 11550) was obtained from the American Type Culture Collection, Rockville, Maryland. Laboratory assays with 1.20 x 10^5 conidia/ml showed slightly lowered chlorophyll a concentrations and dry weight compared to control cultures and a reduction of the number of healthy cells to approximately 50% of the controls. Supernatants of 2-day cultures of this isolate cultured on chitin in Cladophora II medium reduced chlorophyll a concentrations to 50% of control cultures as did A. kiliense supernatants in the same experiment but had no effect on yield or microscopic observations of cell condition. Supernatants from 5-and 9-day cultures had no effect on any of these indicators of cell condition. Additions of 100 and 400 mg/l cephalosporin C (K salt, Sigma Chemical, St. Louis, Missouri) produced some chlorosis, but 10 and 50 mg/l had no effect. It is possible that the toxicant affecting Cladophora is a known compound, but additional studies to determine the identify of the toxic agent must be done.

Many physiological and ecological aspects of the A. kiliense-Cladophora interaction will affect the outcome of any attempt at biological control using the fungus. Similarly use of the toxic agent will require studies of its production and effective concentration on a range of algal species and other organisms. The resistance of various Cladophora strains, or the variable resistance of a given population at different times, as well as the virulence of the fungus culture conditions promoting maximum virulence or highest titer of the toxic agent, host range of the fungus or toxicant and effect of environmental conditions require careful study before any control program could be attempted.

When an endemic pathogen (as opposed to an exotic-introduced pathogen) with narrow host range is used, biological control will result in control of the problem-causing organism. *Acremonium* species are widely distributed in aquatic environments (32, 33, 34, 35). Where a water supply with multiple use is involved, use of a natural agent is preferred over the introduction of a foreign chemical agent.

The positive results in several microcosm experiments with mixed algal communities conducted at different times of the year lend particularly strong support to the reasonableness of further investigating the biological control of *Cladophora* using *A. kiliense* or another fungus. At the end of the August 10-31 microcosm experiment, fish (fathead minnow, *Pimephales promelas*, five per microcosm) were added to one inoculated and one control static microcosm. Every day algae were removed and the water was sieved through a 250 um sieve to collect particles. Algal filaments were removed and fecal pellets were recovered and their dry weight obtained. The weight of fecal matter from animals fed the *A. kiliense* altered algal diet were from 3-13 times ($\bar{x} \pm$ s.d., 6.3 ± 5.1, $n = 4$) greater than those fed the *Cladophora*-dominated original diet. Thus selective control conceivably would increase fish production by fostering development of algal species more desired as food by herbivores than *Cladophora*, thereby promoting more efficient transfer of energy in the food web.

TABLE 1. Viruses, bacteria, and fungi with antagonistic activity against algae (other than *Cladophora*).

Pathogen	Host	Reference
VIRUSES[1]		
A-1 virus	*Anabaena variablis*	Kozyakov, S., B. Gromov, and I. Khudyakov. Mikrobiol. 41:555 (1972).
A-4(L) virus	*Anabaena variablis*	Khudyakov, I. and B. Gromov. Mikrobiol. 42: 904 (1973).
AP-1 virus	*Aphanizomenon flos-aquae*	Granhall, U. Physiol. Plant. 26:332 (1972).
AR-1 virus C-1 virus	*Anabaenopsis raciborskii* *Anabaenopsis circularis* *Raphidiopsis indica* *Cylindrospermum*	Singh, R.N., and P.K. Singh. Nature 216:1020 (1967).
AS-1 virus	*Anacystis nidulans* *Synechococcus cedrorum*	Safferman, R.S., et al. Virol. 47:105 (1972).
LPP-1 virus	*Lyngbya* *Plectonema* *Phormidium*	Safferman, R.S., and M.E. Morris. Sci. 140: 679 (1963).
D-1 virus[2]	*Lyngbya* *Plectonema* *Phormidium*	Daft, M.J., J. Begg, and W.D.P. Stewart. New Phytol. 69:1029 (1970).
G-III virus[2]	*Lyngbya* *Plectonema boryanum* *Phormidium*	Padan, E., M. Shilo, and N. Rislev. Virol. 32:234 (1967).
LPP-2	*Lyngbya* *Plectonema* *Phormidium*	Safferman, R.S., et al. Virol. 39:775 (1969).
LPP-1G virus	*Plectonema boryanum*	Padan, E., et al. Virol. 45:773 (1971).
N-1 virus	*Nostoc muscorum*	Adolph, K.W., and R. Haselkorn. Virol. 46: 200 (1971).
SM-1	*Synechococcus elongatus*	Safferman, R.S. et al. Virol. 37:386 (1969)

TABLE 1. (Continued).

Pathogen	Host	Reference
Unnamed	*Oedogonium*	Pickett-Heaps, J.D. J. Phycol. 8:44 (1972).
Unnamed	*Chlorella pyrenoidosa*	Tikhonenko, A.S., and N.B. Zavarzina. Mikrobiol. 35:850 (1966).
Unnamed	*Sirodotia tenuissima*	Lee, R.E. J. Cell Sci. 8:623 (1971).
BACTERIA[3]		
Bdellovibrio bacteriovorus	*Phormidium* *Mycrocystis*	Burnham, J.C. N.T.I.S. PB-276 569/1WP. 1977.
Cytophaga	Blue-green algae (7 isolates), Green algae (10 isolates)	Stewart, J.R., and R.M. Brown, Jr. Sci. 164:1523 (1969).
Myxobacteria	Blue-green algae	Shilo, M. J. Bacteriol. 104:453 (1970).
Myxobacteria	Blue-green algae (5 species)	Daft, M.J., and W.D.P. Stewart. New Phytol. 70:819 (1971).
Pseudomonas spp.	*Chlamydomonas* *Skeletonema*	Mitchell, R. Nature 230:257 (1971).
Pseudomonas aeruginosa *P. marinoglutinosa*	*Tetraselmis striata* and 12 other species	Berland, B.R., D. J. Bonin, and S.Y. Maestrini. Mar. Biol. 12:189 (1972).
Pseudomonas sp.	*Thallasiosira pseudonanna*	Baker, K.H., and D.S. Herson. Appl. Environ. Microbiol. 35:791 (1978).
FUNGI[3]		
Rhizophidium planktonicum	*Asterionella formosa*	Canter, H.M., and J.W.G. Lund. New Phytol. 47:238 (1948).
Rhizophidium fragilariae *Chytridium versatile*	*Fragilaria crotonesis*	Canter, H.M., and J.W.G. Lund. Trans. Brit. Mycol. Soc. 36:13 (1953).
Chytridium deltanum	*Oocystis* spp.	Masters, M. Canad J. Bot. 49:75 (1971).

TABLE 1. (Continued).

Pathogen	Host	Reference
Rhizophidium androdioctes	*Dictyosphaerium pulchellum*	Canter, H.M. Trans. Brit. Mycol. Soc. 56: 115 (1971).
Zygorhizidium venustum	Chrysophytes	Canter, H.M. Nova Hedwig. 21:577 (1973).
Acremonium *Emericellopsis* *Verticillium*	Blue-green (8) and green (4) algal species	Redhead, K., and S.J.L. Wright. Appl. Environ. Microbiol. 35:962 (1978).
Zygorhizidium melosirae	*Melosira granulata*	Felix, E.A. Trans. Brit. Mycol. Soc. 69: 516 (1977).
Aquatic Phycomycetes	Blue greens, greens, diatoms, desmids	Sparrow, F.K., Jr. Aquatic Phycomycetes. U. Mich. Press. Ann Arbor. 1187 pp. (1960).
Rhizosiphon anabaenae	*Anabaena planktonica*	Patterson, R.A. Ecol. 41:416 (1960).
Rhizophidium sphaerocarpum *Rhizophidium karlingii*	*Spirogyra* *Ulothrix*	Barr, D.J.S., and C.J. Hickman. Can. J. Bot. 45:423 (1967).
Pythium porphyrae	*Porphyra* spp.	Takahashi, M., T. Ichitani, M. Sasuki. Trans. Mycol. Soc. Jap. 18:279 (1977). Arasaki, S. Bull. Jap. Soc. Sci. Fish. 13:74 (1947) Fuller, M. S., B. Lewis, and P. Cook. Mycol. 58:313 (1966).
Lagenidium sp	*Coscinodiscus centralis*	Johnson, T. W., Jr. Mycol. 58:131 (1966).
Lagenisma coscinodisci	*Coscinodiscus granni* *Palmeria hardmania*	Drebes, G. Helg wiss. Meeresunters. 13:426 (1966). Grahame, S. Bri. Phycol. J. 11:57 (1976).

TABLE 1. (Continued).

Pathogen	Host	Reference
Ectrogella perforans *Septalpidium lineare* *Chytridium versatile*	*Synedra*	Johnson, T. W., Jr. Mycol. 58:373 (1966).
Phlyctidium scenedesmi Fott	*Scenedesmus armatus*	Soeder, R. A. and D. Maiweg. Arch. Hydrobiol. 66:48 (1969).
Phlyctidium scenedesmi var. *acuti* var. *nova*	*Scenedesmus acutus*	

1. For reviews see: Safferman, R.S. 1973. Phycoviruses, p. 214-237. *In* N.G. Carr and B.A. Whitton (eds.), The biology of the blue-green algae. U. Calif. Press, Davis. Padun, E. and M. Shilo. 1973. Cyanophages - viruses attacking blue-green algae. Bacteriol. Rev. 37:343-370.

2. May be the same virus as LPP-1

3. For review see Whitton, B.A. 1973. Interactions with other organisms, p. 415-433. *In* N.G. Carr and B.A. Whitton (eds.). The biology of blue-green algae. U. Calif. Press, Davis.

TABLE 2

ASSAY OF EFFECTS OF ISOLATE PR-1 ON C. GLOMERATA IN VARIOUS INCUBATION MEDIA AND IN THE PRESENCE OF ANTIBIOTICS (STREPTOMYCIN SULFATE, FINAL CONCENTRATION 0.06 MG/ML, AND PENICILLIN G, 0.10 MG/ML, 1625 UNITS/MG).

	Control		Inoculated with PR-1	
Culture Conditions	Macroscopic	Microscopic	Macroscopic	Microscopic
White Clay water, autoclaved	Healthy, no apparent contamination	Some chloroplast breakdown, much new growth, no contaminants	Cladophora healthy, fungi growing en mass in Cladophora filaments	Many cells bleached or empty; most damaged cells have fungi associated
White Clay water + antibiotics	Extensive bleaching, extensive fungal development.	Chloroplasts brown, thin hyphae development in and on Cladophora	Same as control with more extensive bleaching	Large mass of thin fungal hyphae associated with or in dead or dying Cladophora
White Clay water	Healthy	Cells-filled with chloroplasts, green	Cladophora green, healthy, slight fungal development	Generally good condition, few empty filaments, some budding, few fungal filaments
Cladophora medium I2	Healthy, no apparent contamination	Cells healthy, much new growth, no contamination	Cells bleached somewhat fungal development	Some cells bleached fungi present
Cladophora medium I2 (25%) + White Clay Water (75%)	Healthy	Healthy, much budding - new growth	Much fungal development along filaments, some bleaching of Cladophora	Large amounts of fungal hyphae

TABLE 3

EFFECT OF *A. KILIENSE* ON *C. GLOMERATA*. RESULTS OF ASSAYS WITH POSITIVE RESULTS.

Experiment	*Cladophora* Source.	*A. kiliense* Inoculum* Range (Conidia/ml in Assay Flask)	Lowest Inoculum Reducing Experimental Treatments to 50% of Control:			*A. kiliense* Culture Conditions
			~~Culture~~ Cell Conditions	Chlorophyll a	Dry Weight	
1	Cultured	$8.32 \times 10^2 - 8.32 \times 10^5$	8.32×10^5	8.32×10^5	8.32×10^5	*Cladophora* mats, 13 days
2	Lititz	$8.32 \times 10^2 - 8.32 \times 10^5$	8.32×10^5	-	8.32×10^5	*Cladophora* mats, 13 days
3	Cultured	$2.08 \times 10^5 - 2.08 \times 10^7$	2.08×10^5	-	-	Ground *Cladophora* in Lititz water, 9 days
4	Cultured	$3.92 \times 10^5 - 3.92 \times 10^7$	3.92×10^6	-	-	Whole *Cladophora* in Lititz water, 9 days
5	Cultured	$2.70 \times 10^4 - 2.70 \times 10^6$	-	2.70×10^5	-	Malt extract broth, 9 days
6	Cultured	$5.12 \times 10^1 - 5.12 \times 10^3$	-	5.12×10^1	-	*Cladophora* Medium II plus chitin, 9 days

* Conidia added in tenfold serial dilution.

TABLE 4

OCCURRENCE AND CONDITION OF *CLADOPHORA* AND *ULOTHRIX* FOLLOWING EXPOSURE TO A. *KILIENSE* IN FLOWING WATER MICROCOSMS, JULY 17-31, 1978. INOCULATED MICROCOSMS RECEIVED ADDITIONS AS IN FIGURE 4.

			Inoculated Microcosms				
			No. Cells Encountered/10^3 Cells	% Cells in Indicated Condition			
				Healthy	Bleached		Empty
Time	Microcosm	Alga			Partially	Chlorotic	
1 wk.	1	*Cladophora*	1000	< 1	3	86	12
	3		1000	50	17	23	10
2 wks.		*Cladophora*					
	1	Attached	600	22	3	60	15
		Surface	20	< 1	0	100	0
	3	Attached	630	21	3	70	6
		Surface	170	65	24	12	< 1
		Ulothrix					
	1	Attached	400	88	12	0	0
		Surface	980	94	5	0	1
	3	Attached	370	43	57	0	0
		Surface	830	89	8	0	2
			Control Microcosms				
1 wk.	2	*Cladophora*	1000	77	9	8	8
	4		1000	71	17	9	4
2 wks.		*Cladophora*					
	2	Attached	970	66	23	8	3
		Surface	170	100	0	0	0
	4	Attached	880	56	19	23	2
		Surface	190	89	< 1	6	6
		Ulothrix					
	2	Attached	30	67	0	0	33
		Surface	830	89	1	8	1
	4	Attached	120	82	0	< 1	18
		Surface	810	78	9	5	9

TABLE 5

OCCURRENCE AND CONDITION OF CLADOPHORA AND ULOTHRIX FOLLOWING EXPOSURE TO A. KILIENSE
IN FLOWING WATER AND STATIC MICROCOSMS, AUGUST 11-31, 1978

			Inoculated Microcosms - Flowing Water						Control Microcosms - Flowing Water				
				% Cells in Indicated Condition						% Cells in Indicated Condition			
					Bleached						Bleached		
Time	Alga	Micro-cosm	No. Cells of Species*	Healthy	Parti-ally	Chlo-rotic	Empty	Micro-cosm	No. Cells of Species*	Healthy	Parti-ally	Chlo-rotic	Empty
8/11	Cladophora	1	1275	98	2	1	0	2	1310	100	0	0	0
		3	1370	97	3	0	0	4	1255	98	2	0	0
	Ulothrix	1	225	100	0	0	0	2	190	100	0	0	0
		3	130	100	0	0	0	4	245	78	22	0	0
8/17	Cladophora	1	490	72	3	14	11	2	1195	92	4	3	1
		3	770	72	3	19	6	4	760	75	2	16	7
	Ulothrix	1	1010	89	10	1	0	2	305	92	8	0	0
		3	730	92	8	0	0	4	740	92	6	0	2
8/22	Cladophora	1	335	3	15	70	12	2	940	38	19	3	40
		2	470	1	0	87	12	4	635	83	5	8	4
	Ulothrix	1	1165	92	8	1	0	2	560	84	14	0	2
		3	1030	97	3	0	0	4	865	86	4	1	9
8/31	Cladophora	1	40	0	0	63	37	2	965	72	18	8	2
		3	145	0	0	62	38	4	415	80	11	7	2
	Ulothrix	1	1460	92	8	1	0	2	535	80	20	0	0
		3	1355	79	20	1	0	4	1085	60	40	0	0
			Inoculated Microcosms - Static						Control Microcosms - Static				
8/11	Cladophora	1	1000	99	1	0	0	2	760	95	4	1	0
		3	790	82	18	0	0	4	810	94	6	0	0
	Ulothrix	1	0	0	0	0	0	2	240	88	12	0	0
		3	210	95	5	0	0	4	190	68	32	0	0
8/22	Cladophora	1	515	25	9	64	2	2	665	84	11	1	4
		3	395	22	9	68	1	4	820	98	1	1	0
	Ulothrix	1	485	41	57	2	0	2	335	99	1	0	0
		3	605	36	60	2	2	4	180	83	17	0	0
8/29	Cladophora	1	135	22	4	67	7	2	745	90	8	1	1
		3	135	15	4	74	7	4	550	94	6	0	0
	Ulothrix	1	865	83	17	0	0	2	255	63	29	2	6
		3	865	93	6	0	1	4	450	81	17	1	1

*1500 cells counted for flowing water microcosms, 1000 cells counted for static water microcosms.

TABLE 6

CONCENTRATION OF SELECTED CHEMICAL PARAMETERS IN LITITZ CREEK WATER ADDED TO MICROCOSMS.

		Concentration (mg/1)	
	N	$\bar{X}$	SD
Total Alkalinity	6	169.0	16.6
NO_3-N	6	8.64	0.47
NO_2-N	6	0.200	0.105
NH_4-N	5	0.083	0.116*
PO_4-P	6	0.295	0.038
pH	6	7.92	0.34

*One Sample was 0.29 mg/1, others ranged from 0.02-0.04 mg/1.

TABLE 7

CONCENTRATIONS OF SELECTED CHEMICAL PARAMETERS IN WATER REMOVED FROM MICROCOSMS, AUGUST 11-31, 1978. MICROCOSMS 1 AND 3 WERE INOCULATED WITH A. KILIENSE: 2 AND 4 WERE CONTROLS.

		Concentration (mg/l) in indicated microcosm							
		Flowing Water				Static			
		1	2	3	4	1	2	3	4
pH	8/18	10.50	10.40	10.45	10.42	10.00	10.60	10.65	10.90
	8/24	10.75	10.72	11.00	10.80				
	8/28	9.75	9.75	10.20	9.65	9.90	10.60	11.00	10.60
	9/1	10.30	10.40	10.45	10.40	10.20	10.20	10.80	10.40
Phenol-phthalein Alkalinity	8/18	44	27	34	28	35	35	48	28
	8/24	36	28	42	28				
	8/28	34	22	43	22	27	25	46	21
	9/1	35	17	31	20	36	23	52	29
Total Alkalinity	8/18	65	48	65	42	93	64	114	66
	8/24	65	62	72	66				
	8/28	131	63	114	73	102	45	83	48
	9/1	73	40	56	43	118	61	112	86
PO_4-P	8/18	0.015	0.015	0.033	0.021	0.913	0.042	0.114	0.051
	8/24	0.158	0.542	0.496	1.020				
	8/28	0.365	0.023	0.096	0.022	0.033	0.010	0.043	0.043
	9/1	0.020	0.068	0.010	0.010	0.265	0.034	0.305	0.170
NO_3-N	8/18	1.82	2.17	2.57	2.27	5.10	4.63	4.42	4.84
	8/24	0.16	0.54	0.50	1.02				
	8/28	0.16	3.79	0.38	4.07	0.20	1.15	1.08	1.86
	9/1	0.23	0.84	0.23	1.78	0.77	3.16	0.23	3.15
NO_2-N	8/18	0.013	0.018	0.015	0.018	0.100	0.095	0.140	0.099
	8/24	0.158	0.542	0.496	1.02				
	8/28	0.006	0.086	0.054	0.074	0.004	0.020	0.051	0.025
	9/1	0.003	0.022	0.003	0.029	0.006	0.062	0.018	0.115
NH_3-N	8/18	0.016	0.015	0.020	0.005	0.085	0.200	0.017	0.023
	8/24	0.021	0.015	0.021	0.021				
	8/28	0.022	0.042	0.060	0.033	0.008	0.011	0.015	0.076
	9/1	0.014	0.001	0.021	0.022	0	0.024	0.004	0

TABLE 8

CONCENTRATION OF SELECTED CHEMICAL PARAMETERS IN LITITZ CREEK WATER AND WATER REMOVED FROM MICROCOSM, OCTOBER 10-20, 1978.

	N	Concentration (mg/l): $\bar{x}$, s.d.				
		Lititz Creek	Microcosm			
			1	2	3	4
Total Alkalinity	2	155.0, 60.1	113.0, 41.0	87.0, 32.5	80.0, 0	102.0, 59.4
NO_3-N	2	6.93, 0.10	5.40, 2.26	5.61, 2.22	4.59, 2.08	5.81, 3.30
NO_2-N	2	0.62, 0.02	0.070, 0.083	0.016, 0.008	0.007, 0.001	0.035, 0.035
NH_4-N	1		0.024	0.024	0.014	0.163
PO_4-P	2	0.649, 0.152	0.190, 0.221	0.059, 0.039	0.022, 0.001	0.088, 0.091
pH	2	8.10, 0.49	9.25, 0.21	9.75, 1.41	9.48, 0.81	9.53, 1.73

TABLE 9

OCCURRENCE AND CONDITION OF *CLADOPHORA* AND *ULOTHRIX* FOLLOWING EXPOSURE TO *A. KILIENSE* IN FLOWING WATER MICROCOSMS, OCTOBER 10-20, 1978.

			Inoculated Microcosms				
				% Cells in Indicated Condition			
			No. Cells of Species*	Healthy	Bleached		Empty
Time	Micro-cosm	Alga			Partially	Chlorotic	
10/10	1	*Cladophora*	1000	91	0	1	9
	3		1000	98	1	0	2
			990	99	0	0	1
	1	*Ulothrix*	0	-	-	-	-
	3		0	-	-	-	-
	4		10	100	0	0	0
10/20	1	*Cladophora*	790	72	4	13	11
	3		840	58	12	11	18
	4		1000	65	7	13	15
	1	*Ulothrix*	210	100	0	0	0
	3		160	97	0	3	0
	4		0	-	-	-	-
			Control Microcosms				
10/10	2	*Cladophora*	990	96	0	1	4
	2	*Ulothrix*	10	100	0	0	0
	2	*Cladophora*	980	95	2	0	3
	2	*Ulothrix*	20	100	0	0	0

*1000 cells counted

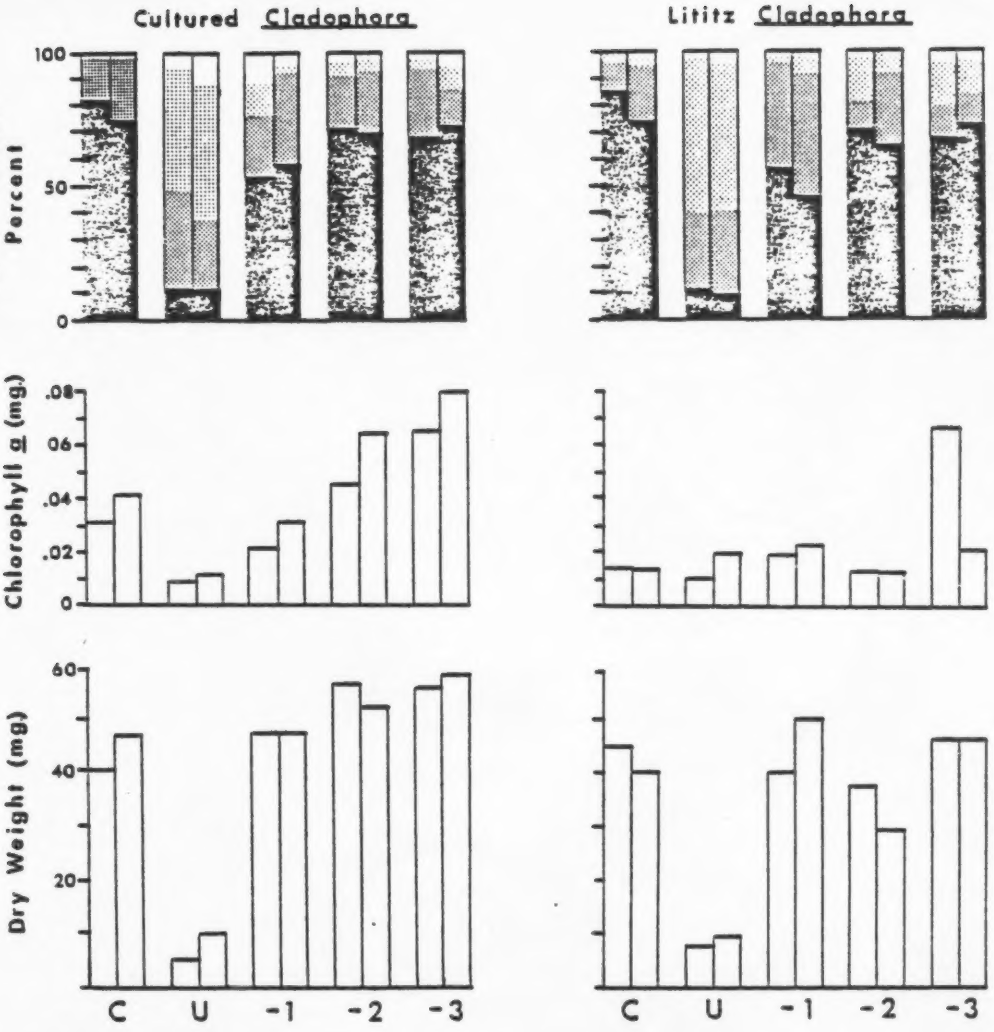

FIGURE 1. Effects of A. *kiliense* (8.32×10^5 conidia/ml U) on cell condition, chlorophyll *a* content and dry weight of *C. glomerata* from stock culture and Lititz Creek. Conidia added in serial dilutions: U=undiluted; -1, -2, -3=tenfold serial dilutions; C=control. Cell condition: Black=healthy, heavy dots=partially bleached, light dots=chlorotic, white=empty.

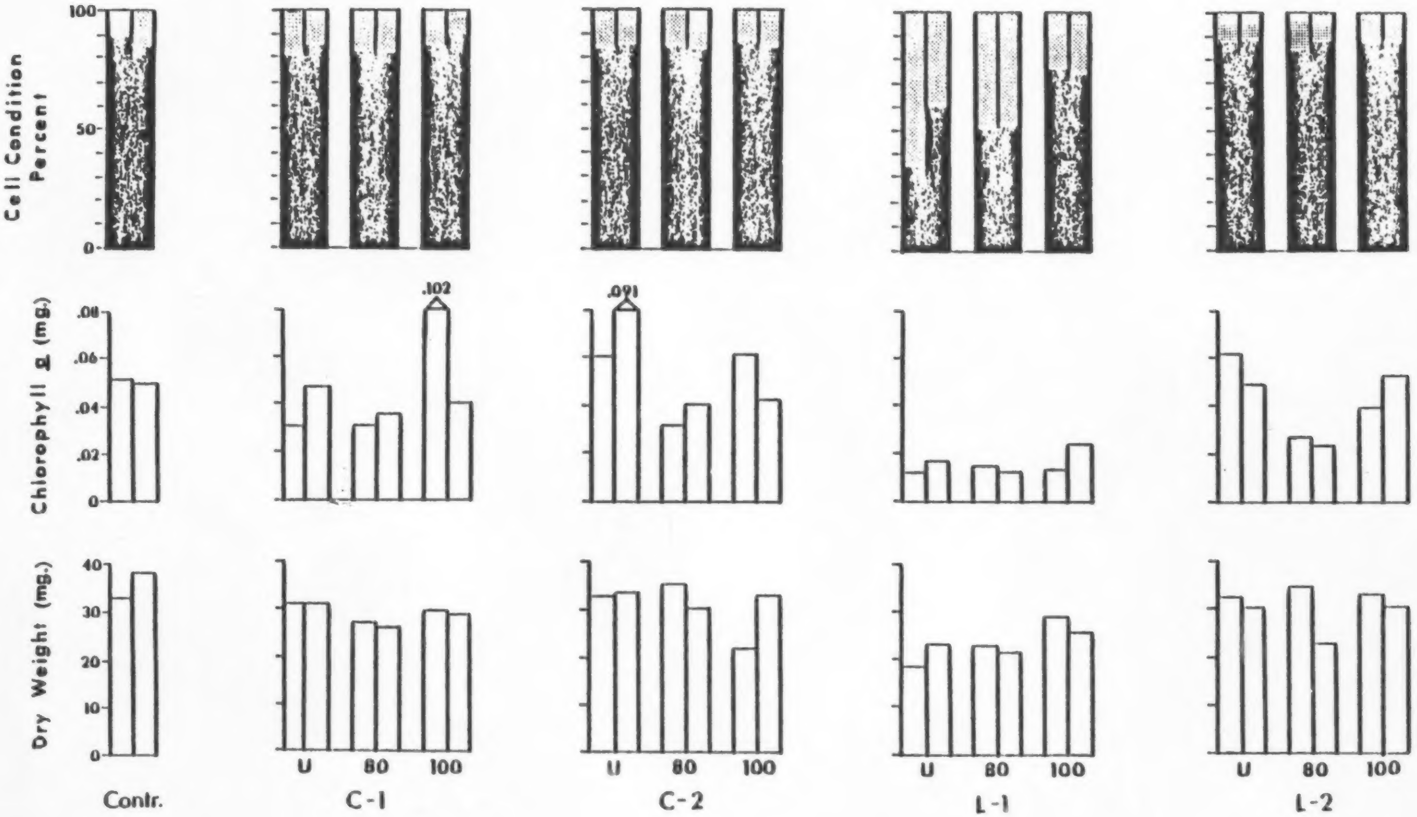

FIGURE 2. Effects of filter-sterilized supernatant fluids from assay flasks in which Cladophora was damaged on C. glomerata from stock culture. Contr.=control; C-1=supernatant from assay with cultured Cladophora receiving undiluted conidia (U), flask 1, in Figure 1 ; C-2=cultured Cladophora, flask 2; L-1=Lititz Cladophora, flask 1; L-2=Lititz Cladophora, flask 2. U=unheated, 80 and 100= heated to 80°C or 100°C for 10 min. before addition. Cell condition: designations as in Figure 1.

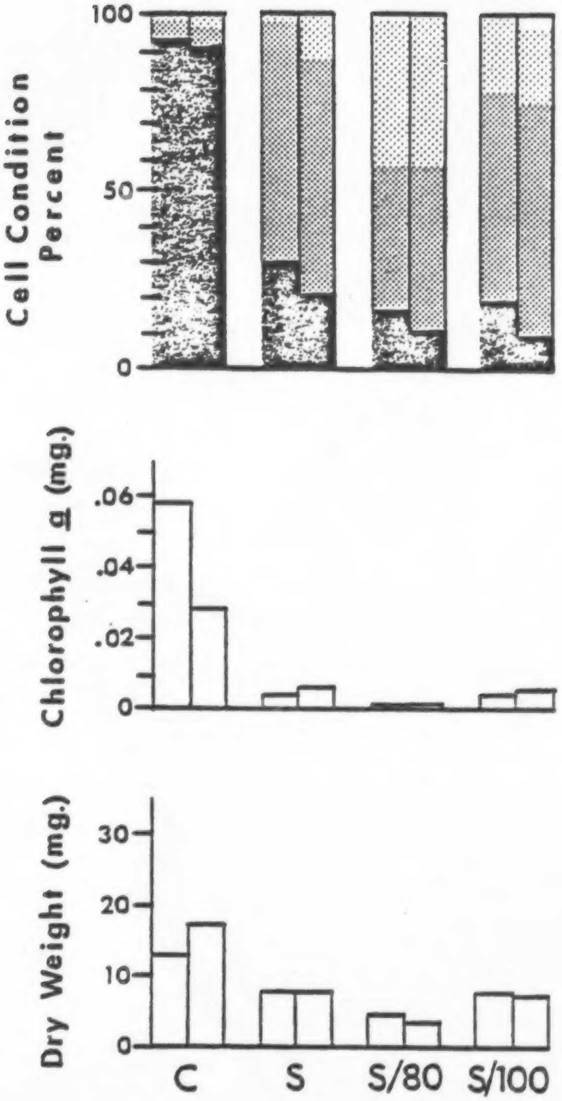

FIGURE 3. Effect of supernatant from *A. kiliense* grown in Czapek-Dox broth at 22°C for 5 days on *C. glomerata* cell condition, chlorophyll *a* content and dry weight. Supernatant filter sterilized before addition. S=supernatant, S/80 or S/100 heated at 80°C or 100°C prior to addition. Control: 5 ml uninoculated broth. Cell condition: designation as in Figure 1.

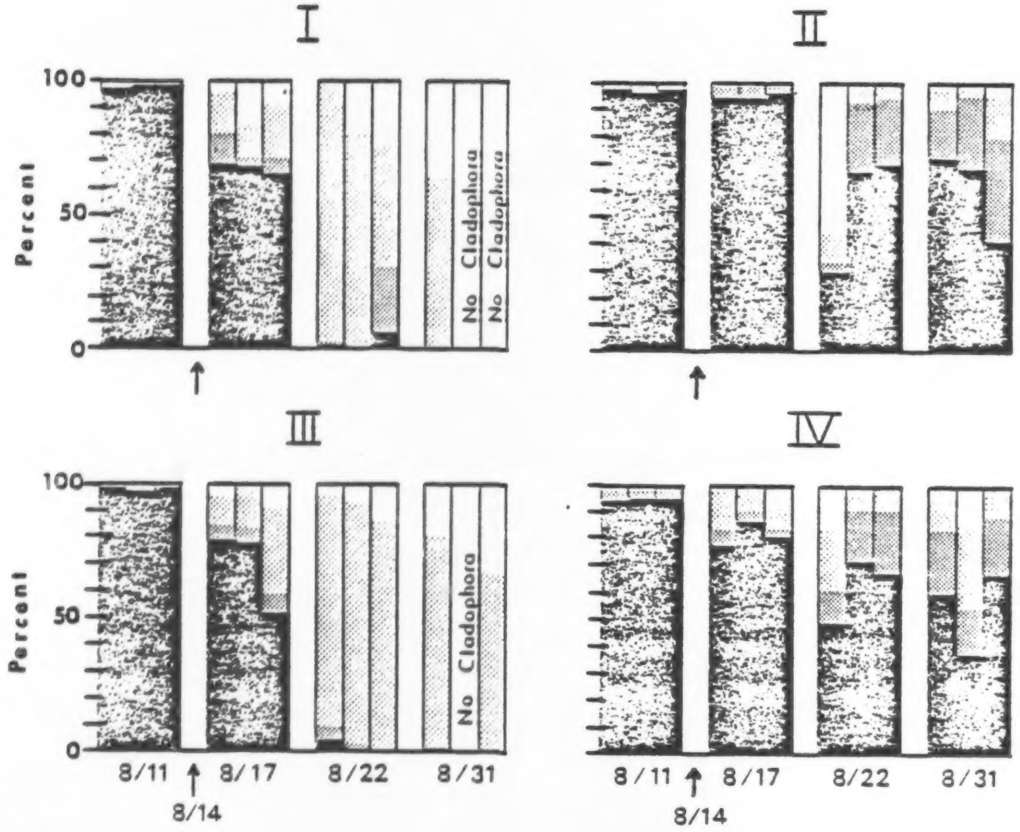

FIGURE 4. Condition of Cladophora cells in flowing water microcosms inoculated with A. kiliense conidia; 113, 6.25, and 29.2 x 10^4/ml on 8/14, 8/17, and 8/21 respectively, and with mycelium and conidia after that (0.32 and 0.39 g wet weight/l to stream I, 0.02 and 0.39 g/l to stream III on 8/24 and 8/28 respectively). Microcosms II and IV=controls. Designations for cell condition as in Figure 1.

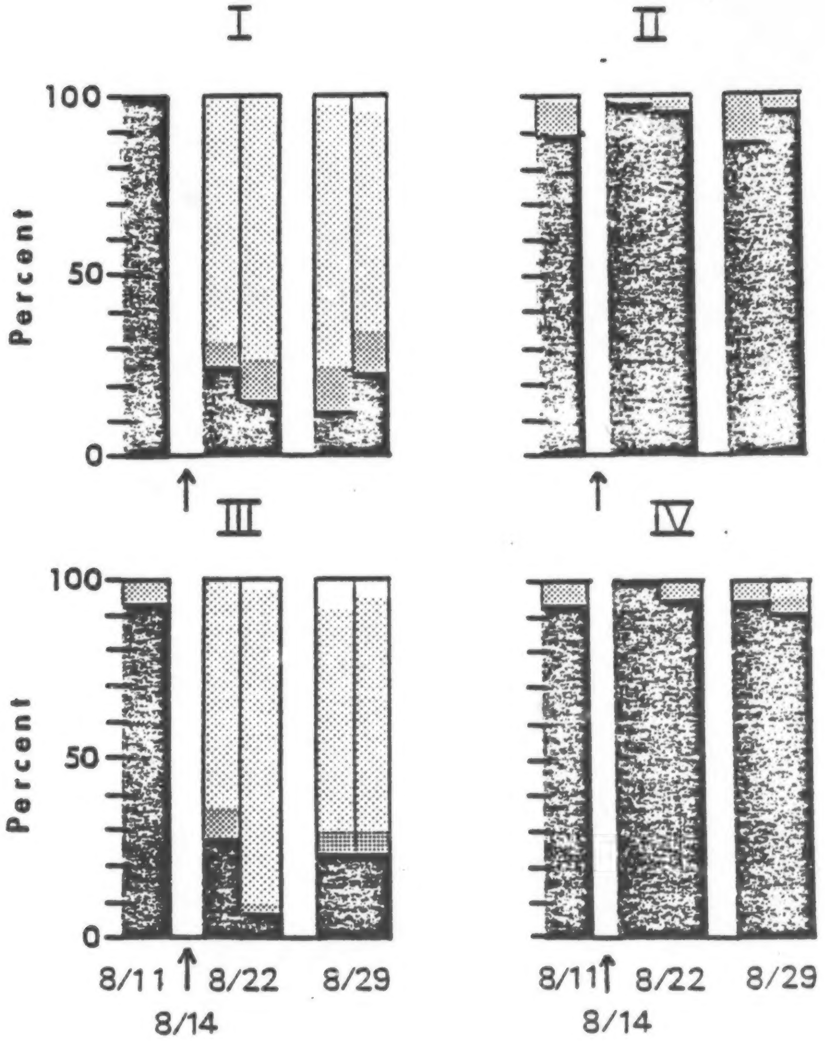

FIGURE 5. Condition of Cladophora cells in static microcosms inoculated with A. kiliense conidia; (113, 6.25, and 28.8 x 10^4/ml on 8/14, 8/17, and 8/21 respectively) and with mycelium and conidia after that (0.64 and 0.78 g wet weight/l to lake I, 0.02 and 0.78 g/l to lake III on 8/24 and 8/28 respectively). Microcosms II and IV=controls. Designations for cell condition as in Figure 1.

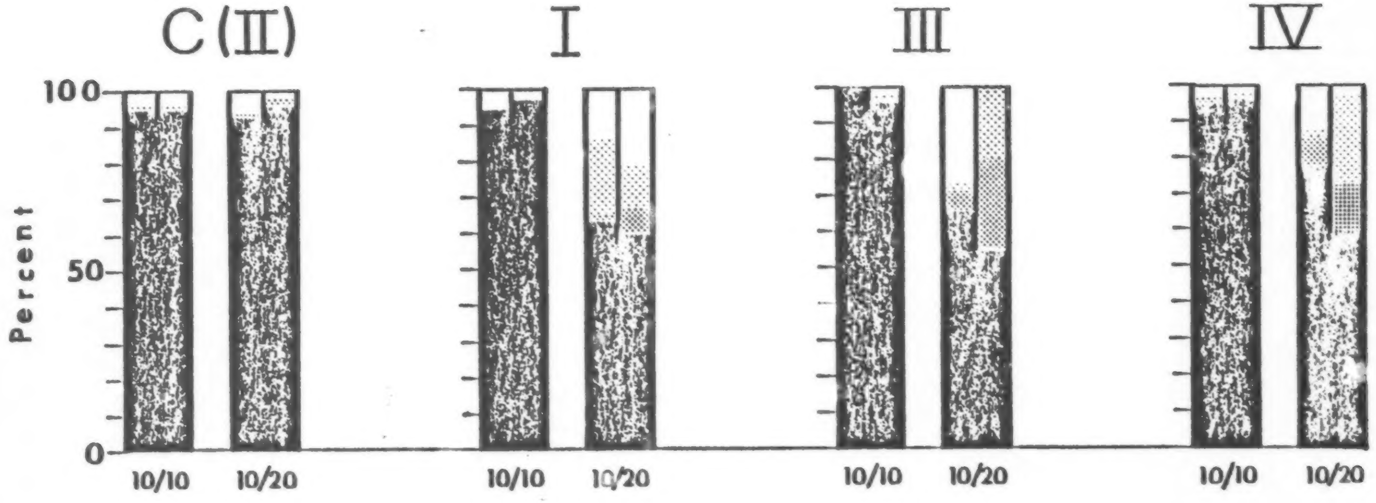

FIGURE 6. Condition of *Cladophora* cells in flowing water microcosms inoculated with *A. kiliense* mycelium and conidia; 0.07-0.21 g wet weight/l added to microcosm I, 0.03-0.10 g/l to microcosm III, 0.01-0.02 g/l to microcosm IV, C (II) = control. 10/10 = sample before exposure, 10/20 = samples after exposure for 10 days. Designation for cell condition as in Figure 1.

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